THE PLANT DISEASE REPORTER

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PLANT DISEASE EPIDEMICS
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IDENTIFICATION SECTION

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The Plant Disease Reporter is issued as a service to plant pathologists throughout the United States. It contains reports, summaries, observations, and comments submitted voluntarily by qualified observers. These reports often are in the form of suggestions, queries, and opinions, frequently purely tentative, offered for consideration or discussion rather than as matters of established fact. In accepting and publishing this material the Plant Disease Epidemics and Identification Section serves merely as an informational clearing house. It does not assume responsibility for the subject matter.

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PLANT DISEASE EPIDEMICS AND IDENTIFICATION SECTION

Horticultural Crops Research Branch — Plant Industry Station, Beltsville, Maryland
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PRELIMINARY STUDIES ON THE STRIGA PARASITIC WEED OF CORN[1]

R. R. Nelson[2]

Abstract

The present knowledge concerning Striga sp., a parasitic flowering plant attacking corn and other gramineous hosts in North Carolina and South Carolina, is summarized. Preliminary surveys have located the weed in 76 infested fields in four counties of southeastern North Carolina and in 38 areas in four South Carolina counties adjoining North Carolina. Host range studies indicate that a wide variety of gramineous species are hosts of the parasite. Early results may suggest differences in the degree of susceptibility of host plants. Percentage germination of Striga seed is increased greatly by periods of dormancy. Seeds of Striga will germinate in the absence of host stimuli but soon die, indicating a need for substances excreted by host roots. Moisture and temperature are important factors in the germination of Striga seed and the subsequent development of the plant. Tentative suggestions for the partial control and containment of Striga are included.

INTRODUCTION

There are many species of parasitic flowering plants that attack a wide variety of cultivated and wild hosts throughout the world. Within the genus Striga there are a number of species that parasitize such important economic hosts as corn, sugarcane, tobacco, rice, and some of the small grains (3, 4, 6, 7). Species of Striga occur commonly in tropical and sub-tropical areas of the Eastern Hemisphere and are known to be serious parasites in Africa, Australia, India, and Indonesia. The recent discovery of a species of Striga parasitic on the roots of corn and gramineous weeds in North Carolina (2), and subsequently in South Carolina, is the first time this plant has been identified in the Western Hemsiphere. The species had not been positively identified at time of writing, but taxonomists report that the plant resembles S. asiatica and also is similar to S. lutea, a species known to parasitize corn in South Africa (6)[3]. Botanically, the species is characterized by orange to red flowers, square aerial stems becoming rounded below ground, alternate leaves and minute striated seed borne in capsules (Fig. 1 to 4). For the purpose of this report the parasite is referred to simply as Striga.

DISTRIBUTION

Very little is known about the prevalence and geographic distribution of the weed in North Carolina and the surrounding area. Striga was not identified as occurring in North Carolina until mid-July 1956. Preliminary surveys conducted by the United States Department of Agriculture and State agencies in the late summer and early fall of 1956 indicate that the species is present in at least four counties in southeastern North Carolina and in the four adjoining counties in northeastern South Carolina. These counties with the number of known infested areas are: North Carolina: Columbus, 40; Robeson, 23; Bladen, 8; and Cumberland, 5. South Carolina: Dillon, 17; Horry, 16; Marion, 4; and Marlboro, 1. Striga has been observed

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[1] Cooperative investigations of the Field Crops Research Branch, Agricultural Research Service, United States Department of Agriculture and the North Carolina Agricultural Experiment Station.

[2] Plant Pathologist, Crops Research Division, Agricultural Research Service, United States Department of Agriculture.

[3] [Dr. S. F. Blake has identified it as S. asiatica -- Ed. Note.]

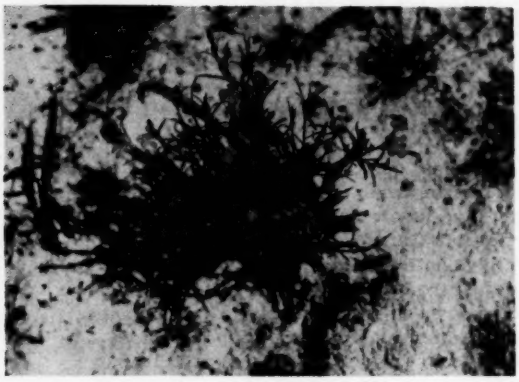

FIGURE 1. Striga sp. in field. Note size and compactness of plant.

FIGURE 2. Striga sp. parasitic on corn in greenhouse. Note alternate leaves.

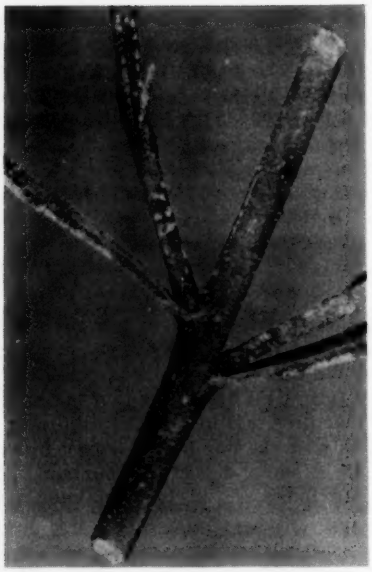

FIGURE 3. Square aerial stem of Striga sp.

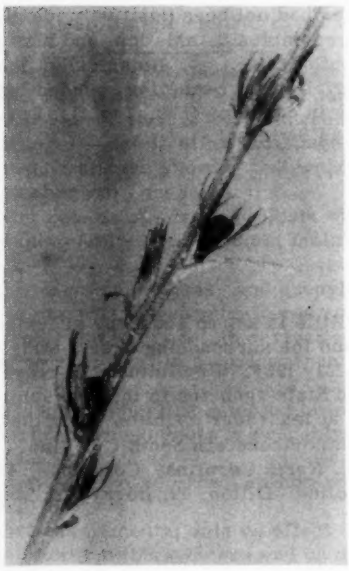

FIGURE 4. Seed capsules of Striga sp.

in many fields of corn and in one field of milo. It has also been observed parasitizing crabgrass in ditches and along road-sides, and attacking this host and probably other grasses in fields planted to other crops. Although the parasite has been observed on gramineous weeds in fields of tobacco, peanuts, cotton, soybean, cowpea, and other non-gramineous crops, it apparently does not attack those crops. The complete distribution and prevalence of Striga are still not known. Survey officials indicate that even within the limits of the known area of infestation, only 20 percent of that area has been surveyed. Additional surveys undoubtedly will supplement the present information.

MODE OF PARASITISM

Striga is an obligate parasite which attacks the roots of susceptible plants. If a corn plant and associated Striga plants are dug up and the soil carefully washed away, one can readily distinguish the white roots of the parasite from the darker brownish roots of the corn plant. Both sets of roots are heavily intertwined (Fig. 5), and on careful examination it can be seen that the roots of the parasite are attached to the roots of the host by means of small bell-shaped swellings, similar in appearance to appressoria (Fig. 6-7). The true mechanism of parasitism for the species is not known.

Striga plants are relatively small, usually from 6 to 12 inches in height and seldom exceeding 18 inches. Frequently the weed can be found at some distance removed from the base of the corn stalk. In such instances it might be attacking the roots of corn or possibly parasitizing a grass weed. Within an infested area, Striga may be observed at various stages of growth, ranging from small seedlings to mature plants bearing seed. Unless an observer were aware of the parasitic nature of Striga he would likely fail to associate the weed with retarded growth and drying of corn.

Information is not available at the present time concerning when, where, and how the initial infestation occurred. Statements by a number of farmers in the infested areas tentatively place the earliest observed infestation between 1946 and 1950.

RESULTS OF EXPERIMENTAL INVESTIGATION

Host Range: — Seventy-two plant species including cultivated and weed plants common to the region have been incorporated in preliminary host range studies in the greenhouse. Thirty-two of the species were grasses representing 26 genera. The 30 non-gramineous species belonged to 13 families in the Dicotyledoneae. Each species was tested in soil obtained from infested fields and in sterilized soil to which freshly harvested seed of Striga was added at the time of planting. Since Striga is an obligate parasite, the appearance of the weed above the soil was taken to indicate that the plant species in question was susceptible.

Table 1. Plant species susceptible to the species of Striga present in North Carolina, as determined in studies to date.

Common name	Latin name	Number of days needed for Striga to appear	
		Least	Range
Corn	Zea mays L.	30	30-114
Sorghum	Sorghum vulgare Pers.	28	28-66
Sweet Sudan	S. vulgare var. sudanense (Piper) Hitchc.	34	34-77
Carpet grass	Axonopus affinis Chase	61	61-82
Crab grass	Digitaria sanguinalis (L.) Scop.	49	49-67
Johnson grass	Sorghum halepense (L.) Pers.	29	29-71
Sandbur	Cenchrus sp.	74	No data
Foxtail millet	Setaria italica (L.) Beauv.	73	73-108
Barnyard grass	Echinochloa crusgalli (L.) Beauv.	78	78-98
Japanese millet	E. frumentacea	53	53-80
Goose-grass	Eleusine sp.	31	31-63
Rescue-grass	Bromus catharticus Vahl	48	No data

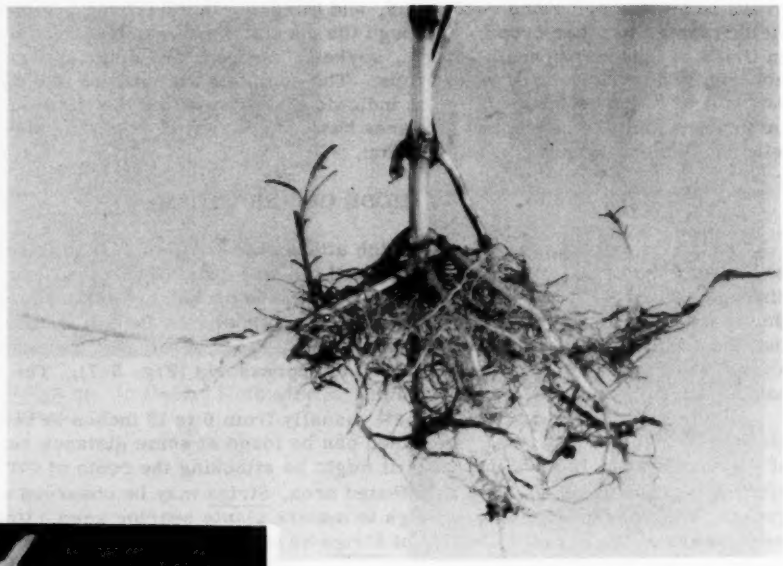

FIGURE 5. Roots of Striga sp. plants intertwined with corn roots.

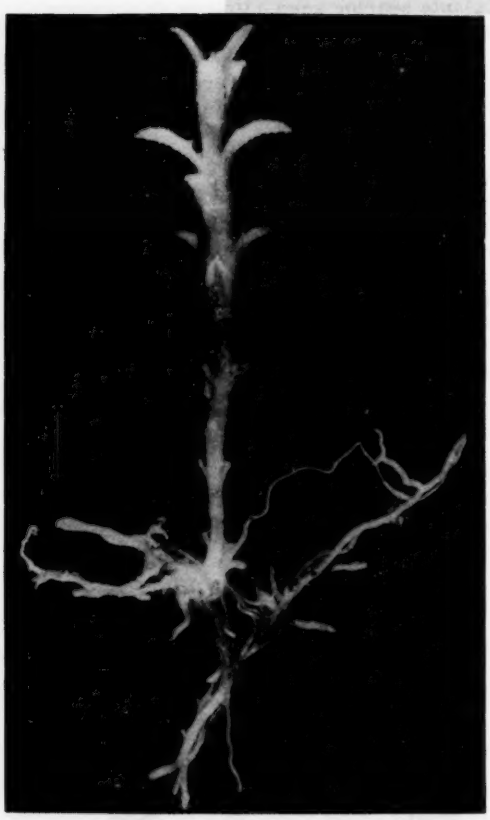

FIGURE 6. Immature plant of Striga sp. attached to corn root.

FIGURE 7. Attachments of roots of Striga sp. to corn roots showing bell-shaped structures.

To date 13 species in 10 genera have been found to be susceptible to Striga (Table 1). None of the non-gramineous species has been shown to be susceptible. The work of Saunders (6) with S. lutea indicated that only grass hosts were susceptible to that species.

The failure of the weed to appear aboveground does not necessarily imply that a plant species is resistant. Germinating seed of orchard grass (Dactylis glomerata L.) and Bermuda grass (Cynodon dactylon (L.) Pers.) have stimulated a small percentage of Striga seed to germinate in the laboratory although Striga has not appeared in pots of those grasses in the greenhouse. Sandbur (Cenchrus sp.) and Dallis grass (Paspalum dilatatum Poir), however, were shown to be susceptible in the greenhouse, but preliminary efforts to induce Striga seed to germinate in their presence in the laboratory failed.

Striga appears to be a very sensitive plant and any number of factors probably are capable of inhibiting or retarding its development. The length of time required for plants of Striga to appear aboveground has varied tremendously, not only among the susceptible hosts but also for a single suscept during repeated tests. Table 1 lists the susceptible species and the time required for Striga to appear. Such a differential in time makes it difficult to determine whether a species is resistant or whether some factor has prevented true expression of parasitism. Nevertheless, these results may suggest a distinct difference in susceptibility among host species as measured by time required for Striga to emerge.

Studies on Germination: -- The research of investigators in other countries indicated that seed of species of Striga require a rather prolonged period of dormancy prior to germination. In the case of Striga lutea, for example, maximum germination is attained 18 months after the seed has matured (6). Present greenhouse and laboratory studies in North Carolina indicate that at least a small percentage of seed of Striga sp. will germinate without a dormancy period. Freshly harvested seed was placed in sterile soil planted to corn. In 6 weeks several Striga plants appeared in the pots of corn. Additional seed of the same lot germinated within 10 days at 32° C when placed on moistened filter paper in a Petri plate containing germinating corn seed. These studies further indicated that periods of dormancy increase the percentage of germination. Lots of seed harvested in the fall of 1956 were tested for germination at 4-week intervals. In all cases the percentage germination increased with each time increment.

It has been reported that seed of Striga spp. will germinate only in the presence of a substance which apparently is excreted by host roots, although some workers have shown that no stimulus is needed (5, 6). In the present tests, a small percentage of seed germinated in the absence of any host or host excretions. In a series of treatments including scarification and alternate freezing and thawing, from 3 to 9 percent germination was attained. Best results were obtained when scarified seed were frozen and thawed while dry rather than wet, and scarification was the one common denominator in all variations of the studies. Without exception, the germinated seed ceased development within 24 hours and presumably died.

Effect of Moisture on Germination and Growth: -- Moisture appears to be an important factor in seed germination and in the subsequent development of the plant. Preliminary laboratory studies indicate that seed will not germinate in the presence of free water, even when in direct contact with roots of susceptible hosts. In the greenhouse the growth and development of Striga was inhibited in saturated soils. Three pots each of corn, sorghum, and Johnson grass, planted in infested soil, were watered in the usual fashion, while two pots of the same hosts were kept saturated by continual watering. Striga developed normally in the lightly watered pots, while no Striga appeared in pots watered excessively. When the soil of the latter was carefully washed away, no young immature Striga plants were observed, indicating that germination of Striga seed had not occurred. A third set of duplicate pots of the aforementioned hosts was watered normally for four weeks, presumably sufficient time for Striga to germinate and make parasitic contact. After four weeks, all pots were watered excessively. No Striga plants appeared by the end of 10 weeks. When the soil was carefully washed away, young seedlings of Striga were readily observed, but apparently were inhibited or retarded.

Effect of Temperature on Germination and Growth:-- Temperature is an equally important factor influencing seed germination and plant growth. The effect of temperature on germination of seed was studied in a series of laboratory tests. Striga seed were placed in Petri plates on filter paper moistened with water that had been percolated through pots of vigorously growing corn plants. The leachate presumably contained the substance or substances excreted by the roots of corn that induce the seed to germinate. The seed were incubated for 3 weeks at 4-

degree intervals ranging from 8° to 40° C. The results are summarized in Table 2. The studies indicated that maximum germination is attained at approximately 32° C, a comparatively high optimum temperature. It is further indicated that high temperatures not only favor a higher percentage germination, but also that seed have a tendency to germinate more rapidly at the higher temperatures. The minimum temperature required for germination in these tests was approximately 20° C, although appreciable germination did occur below 24° C. The maximum temperature was not established, although some germination occurred at 40° C.

Table 2. Effect of temperature on germination of Striga seed.

Temperature (°C)	Percent germination after 3 weeks[a]	Number of days required for initial germination
8	0	—
12	0	—
16	0	—
20	.75	17
24	22.5	13
28	47.0	7
32	53.5	6
34	42.8	6
36	39.5	6
40	29.8	5

[a]Based on a total of 400 seeds at each temperature.

While the comparisons of percentage germination at the various temperatures employed are probably accurate, the actual percentages may not be entirely indicative. The seed used had been dormant for approximately 5 months prior to these studies. If earlier reports are indicative, a considerably longer period is required for maximum germination.

SUGGESTED CONTROL MEASURES

Agricultural workers representing the United States Department of Agriculture and agricultural agencies of North Carolina and South Carolina met in December 1956 to discuss problems relative to Striga. While it was recognized that more research is needed, the following recommendations, based on present knowledge, were made in an effort to reduce the prevalence of Striga within infested areas and to minimize the possibility of its spread:

1. Discontinue planting corn and sorghum on infested land.
2. Adopt rotation practices.
   (a) Plant small grains in the fall following recommended practices.
   (b) In late spring or early summer follow with "trap" or "catch" crops as described in "3" below.
3. Summer planted crops may be replaced with "catch"* crops which should be planted and turned under before Striga flowers or by planting "trap"* crops.
   * A "catch" crop is one on whose roots Striga seed will germinate and grow to produce seed. Planting and turning these crops should be continued as necessary throughout the entire summer growing period. Sudan grass serves for this purpose.
   * A "trap" crop is one on whose roots Striga seed will germinate but the weed fails to grow. Cowpeas and soybeans serve this purpose. These crops should be harvested only on farms where the Striga infestation is severe and the harvested crop is to be used in the infested area or on the individual farm.
4. Such host plants as crabgrass and other grasses remaining in the field should be thoroughly destroyed by plowing or by the use of effective herbicides.

5. Non-cultivated lands in which Striga appears should be treated by chemical or cultural methods to destroy the pest in the field before flowering.
6. Cultivated crops in which Striga is found for the first time should be treated by chemical or cultural methods to destroy the pest in the field before flowering.
7. Wherever Striga is suspected do not move plants from the area, but request on-the-farm-identification through your county agent.
8. As far as known, broadleaf crops such as cotton, tobacco, peanuts, and sweetpotatoes may be grown on Striga-infested land. But, such crops must be kept free of host plants, such as crabgrass.
9. All precautions should be used to prevent movement of Striga plants or seeds through transplanted crops, soil, by machinery, or any other means.

Further research is in progress.

Literature Cited

1. FULLER, C. 1900. Rooi-bloem or witchweed. First Rept. Gov. Entom. Natal, 1899-1900; pp. 20-22.
2. GARRISS, HOWARD R., and J. C. WELLS. 1956. Parasitic herbaceous annual associated with corn disease in North Carolina. Plant Dis. Reptr. 40: 837-839.
3. LUTHRA, J. C. 1921. Striga as a root parasite of sugarcane. Agric. Jour. India 16: Pt. 5.
4. PALM, B. T., and C. HEUSER. 1924. Striga lutea als Schädiger des Reises auf Sumatra. Zeitsch. für Pflanzenkrankh. 34: 11-18.
5. PEARSON, H. H. W. 1913. The problem of the witch-weed. Dept. Agric., Union South Africa Bull. 40: 3-34.
6. SAUNDERS, A. R. 1933. Studies on phanerogamic parasitism with particular reference to Striga lutea. South African Dept. of Agric. Science Bull. 128.
7. WILD, H. 1948. A suggestion for the control of tobacco witchweed by leguminous trap crops. Rhod. Agric. Jour. 45: 208-215.

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CEPHALOSPORIUM LEAF STRIPE DISEASE OF WINTER WHEAT

D. J. Tyler and L. E. Dickens

Winter wheat plants, at the heading stage, which manifested stripe symptoms of disease on leaf blades and sheaths were collected from central and western New York fields in June 1956. Diseased tissue from leaves and sheaths yielded macroscopically similar fungous colonies when placed on potato dextrose agar in Petri dishes. Colonies of the same fungus were obtained from stripe-diseased specimens of wheat collected in widely separated New York fields. The fungus appears to be the same as that which causes a stripe disease of wheat in Japan and which Y. Nisikado found in Japan for the first time in May 1932, and named Cephalosporium gramineum Nisikado & Ikata (Ohara Inst. F. Landw. Forsch. Ber. 6: 275-306. 1934). G. W. Bruehl reported the presence of wheat stripe, caused by Cephalosporium gramineum Nisikado & Ikata, in the State of Washington, U.S.A., in 1956 (Phytopath. 46: 178-180).

Pathogenicity of the fungus isolated from diseased wheat plants found in New York fields was proved as follows: A purified sub-culture of the fungus isolated from striped wheat plants was planted in potato dextrose nutrient solution where conidia developed abundantly at temperatures ranging from 70° to 75° F. Conidia that formed in the nutrient solution during a growth period of 8 days were recovered by centrifugation and they were washed twice in sterile distilled water. The washed conidia were suspended in sterile water and inoculated into plants of winter wheat (Cornell 595), winter barley (Hudson), and oats (Garry and Dubois). The plants were grown in a greenhouse and inoculum was injected, by means of a hypodermic syringe, into the leaf whorls at the growing points immediately prior to elongation of the lowermost internode. Stripe symptoms appeared on the leaves and sheaths of the three kinds of cereal plants two to three weeks after they were inoculated. The symptoms on wheat were similar to those described by Nisikado et al., and by Bruehl (loc. cit.). Isolates of the fungus were obtained from artificially inoculated plants of wheat, barley, and oats that developed stripe symptoms. These isolates were morphologically identical to the fungus that was isolated originally from the wheat plants affected by the stripe disease and which were collected in New York fields. Conidia of the Cephalosporium that caused stripe of wheat in New York, averaged 2.21 μ in width by 6.30 μ in length, and they ranged in size from 1.4 μ to 2.8 μ in width by 4.2 μ to 9.2 μ in length. Colonies of this fungus grew slowly on potato dextrose agar at temperatures of 70° to 80° F, and they were not pigmented. Size of the conidia, growth characteristics of the fungus on potato dextrose agar, and symptoms induced in wheat by the New York fungus leads to the conclusion that Cephalosporium gramineum Nisikado & Ikata is the causal agent of a stripe disease of winter wheat observed in New York. A survey is needed to determine whether C. gramineum naturally attacks barley and oats, or other Gramineae, in New York.

The exact range of the causal agent of Cephalosporium stripe of winter wheat in the State is not known, but it has been observed in all of the important soft white winter wheat growing counties of central and western New York. The known range of the pathogen in New York indicates, therefore, that its establishment here was not accomplished in recent years.

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SEPTORIA REACTIONS OF OAT VARIETIES AND SELECTIONS IN 1956[1]

A. L. Hooker[2]

Abstract

The reactions of 257 oat varieties or selections to artificial inoculations with Septoria avenae were determined at Madison, Wisconsin in 1956. Significant differences in varietal reaction were measured in percentage of leaf infection, in leaf lesion size, in percentage of stem infection, and in percentage of kernel infection. Several oat strains showed little disease while others were highly susceptible.

Septoria avenae Frank infects the leaves, stems, and kernels of the oat plant, producing characteristic symptoms and reducing grain yield and quality. Oat varieties and strains differ in the degree of leaf, stem, and kernel infection. The reaction of 153 oat varieties or selections to artificial inoculations in 1955 were summarized in a previous report.[3] The present paper summarizes the results of a similar test with 257 varieties or selections conducted in 1956.

MATERIALS AND METHODS

A majority of the established varieties and advanced experimental selections of oats adapted to the North Central and Northeastern States were artificially inoculated with Septoria avenae in a disease nursery at Madison, Wisconsin, in 1956. Three replications of the 257 oat strains were sown on April 19 in 3-foot rows in a randomized complete block design. All plots were inoculated by spraying and by hypodermic injections following methods similar to those employed in 1955[3].

Leaf infection scores were recorded as percentage of leaf area infected on a whole plant basis, and ratings for leaf lesion size, ranging from 1 (small and restricted) to 5 (large and spreading) were taken on June 28. Stem and kernel infection scores were recorded at maturity. Ten culms from each plot, in addition to those inoculated hypodermically, were taken into the laboratory and rated as to the percentage of culm area infected after the leaves and leaf sheaths had been removed. The seed from the hypodermically inoculated culms was rated for percentage of kernel infection.

Statistical analyses were calculated, using all 771 entries in the test.

RESULTS

The oat varieties differed widely in their reaction to the Septoria disease (Table 1). Statistically significant differences in infection of the leaves, stems, and kernels were recorded (Table 2). The data on stem infection following hypodermic inoculations are not presented in Table 1 because the ratings closely paralleled those from the spray inoculations.

The Clintafe, Clintland, Clinton, Clinton 59, Gopher, and Vicland varieties showed the least infection by Septoria avenae in the 2-year tests and the Andrew, Bentland, Clarion, Fayette, Logan, Newton, and Rodney varieties showed the most infection. Of the varieties tested only in 1956, Columbia, Craig, Dupre, Kanota, Mohawk, Roxton, and Victory had low Septoria ratings while Bond appeared to be very susceptible.

In addition to the named varieties, over 200 experimental selections were evaluated for Septoria reaction. Selections of the following crosses had low or high Septoria leaf and stem infection in 1956:

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[1]Cooperative investigation between the Crops Research Division, Agricultural Research Service, United States Department of Agriculture, and the Wisconsin Agricultural Experiment Station. Published with the approval of the Director of the Wisconsin Agricultural Experiment Station.

[2] Plant Pathologist, Crops Research Division, Agricultural Research Service, United States Department of Agriculture, Madison, Wisconsin. The author is indebted to R. W. Ruhde for technical assistance.

[3]Hooker, A. L. 1955. Septoria reactions of entries in the North Central States uniform oat yield nursery and other strains in 1955. Plant Dis. Reptr. 39:963-966.

Table 1. Leaf, stem, and kernel reactions to Septoria avenae of 40 oat varieties at Madison, Wisconsin, in 1956. Leaf and stem ratings followed spray inoculations and kernel ratings followed hypodermic inoculations. Average of 3 replications.

Variety	C.I. No.[a]	Leaf lesion size[b]	Percentage of infection: 1956 tests			Two-year average		
			Leaf	Stem	Kernel	Leaf	Stem	Kernel
Ajax	4157	2.3	28	25	5	28	24	3
Andrew	4170	2.7	42	58	7	40	55	4
Beedee	6752	2.3	28	18	25	33	18	26
Bentland	6930	3.3	37	52	20	44	48	17
Bond	2733	3.3	40	55	7	—	—	—
Branch	5013	2.7	30	17	11	30	21	13
Burnett	6537	4.3	38	18	2	39	30	6
Cherokee	5444	3.3	38	32	5	40	34	4
Cherokee Reselect	7194	3.3	50	70	8	—	—	—
Clarion	5647	3.3	38	58	23	40	62	14
Clintafe	5869	3.3	37	12	9	32	14	6
Clintland	6701	2.3	28	20	22	30	12	12
Clinton	3971	3.3	35	7	8	34	7	4
Clinton 59	4259	3.0	28	7	17	34	6	12
Columbia	2820	3.0	38	12	2	—	—	—
Craig	5332	3.0	35	10	3	—	—	—
Dupre	4372	3.3	37	12	2	—	—	—
Early Clinton	7192	3.7	42	32	3	—	—	—
Fayette	6916	3.7	43	60	45	41	55	36
Garry	6662	3.3	42	40	8	37	35	6
Gopher	2027	3.3	38	8	2	36	6	2
Jackson	5441	3.7	38	12	3	36	22	2
Kanota	839	3.3	30	10	12	—	—	—
Logan	6929	3.0	43	45	8	44	54	10
Minland	6765	2.3	37	30	16	44	35	22
Mo. 0-205	4988	3.0	35	27	25	36	37	18
Mohawk	4327	3.0	33	5	1	—	—	—
Nemaha	4301	3.0	40	38	15	38	33	18
Newton	6642	3.0	38	57	15	39	54	16
Osage	3991	3.3	35	27	9	—	—	—
Putnam	6927	3.3	42	20	37	43	30	34
Rodney	6661	3.3	35	50	7	38	52	10
Roxton	4134	3.3	35	13	1	—	—	—
Sauk	5946	3.0	32	20	8	34	32	15
Scotian	7203	2.7	35	22	1	—	—	—
Simcoe	6767	2.3	30	18	15	31	20	14
State Pride	1154	3.0	38	23	2	38	24	3
Tama	3502	2.7	35	17	20	—	—	—
Vicland	3611	2.7	38	13	4	38	10	12
Victory	560	3.7	37	5	5	—	—	—

[a] Refers to accession number of Cereal Crops Section, Crops Research Division
[b] Range 1 (small and restricted) to 5 (large and spreading)

Low Leaf Infection (30% or Less)

Ajax x (Hawkeye x Victoria)
Clintland x (Garry x Hawkeye-Victoria)
Clinton x (Hawkeye x Victoria)
Clinton^4 x Santa Fe
(Clinton^2 x Santa Fe) x (Hawkeye x Victoria)
Clinton x (Victory x (Victoria x Hajira-Banner)
(D69 x Bond) x (Victoria-Richland x Bannock)
Forvic x (Hawkeye x Victoria)
Forward^2 x (Victoria x Richland)
(Landhafer x (Mindo x Hajira-Joanette)) x Andrew
(Landhafer x (Mindo x Hajira-Joanette)) x Clinton
Vicland x Erban
Vicland x (Landhafer x Richland-Bond)
(Victory x Hajira) x (D69 x Bond)

High Leaf Infection (45% or More)

Andrew x Landhafer
(Bonda x (Hawkeye x Victoria)) x (Garry x (Hawkeye x Victoria))
Bonda x (Landhafer x (Forward x Victoria-Richland))
Erban x (Richland x Bond)
(Landhafer x (Bond-Rainbow x Hajira-Joanette)) x Clinton
(Landhafer x (Mindo x Hajira-Joanette)) x Andrew
Nemaha x (Neosho x Landhafer)
Osage x ((Bonda x Hajira-Joanette) x Santa Fe)
Rodney x (Landhafer x (Forward x Victoria-Richland))
Vicland x (Branch x Clinton^2-Santa Fe)

Low Stem Infection (15% or Less)

Clinton^4 x Santa Fe
Clinton x (Victory x (Victoria x Hajira-Banner))
(D69 x Bond) x (Victoria-Richland x Bannock)
Erban x Boone
((Forward x Victoria-Richland) x Vicland) x (Hawkeye x Victoria)
Goldwin x Boone
Goldwin x Clinton
(Hawkeye x Victoria) x Forvic
(Landhafer x (Bond-Rainbow x Hajira-Joanette)) x Andrew
(Landhafer x (Bond-Rainbow x Hajira-Joanette)) x Clinton
(Landhafer x (Mindo x Hajira-Joanette)) x Clinton
Landhafer x (Richland x Bond)
Rodney x (Santa Fe x Clinton^3)
Vicland x Erban
(Victory x Hajira) x (D69 x Bond)

High Stem Infection (50% or More)

Andrew x Landhafer
((Anthony x Bond) x Boone) x ((Bond x Rainbow) x (Hajira x Joanette))
(Clarion x Bonda) x (Hajira-Joanette x Santa Fe)
Columbia x Marion
(Columbia x Victoria-Richland) x Mindo
(Landhafer x (Bond-Rainbow x Hajira-Joanette)) x Andrew^2
(Landhafer x (Bond-Rainbow x Hajira-Joanette)) x Clinton
(Landhafer x (Mindo x Hajira-Joanette)) x Andrew
Marion x (D69 x Bond)
Osage x ((Bonda x Hajira-Joanette) x Santa Fe)
Rodney x (Santa Fe x Clinton^3)
Vicland x (Landhafer x (Richland x Bond))

Table 2. Analyses of variance summary for reaction of 257 oat varieties or selections to Septoria avenae.

Disease evaluation	MS	F	LSD .05
Leaf lesion size	0.71	1.71**	1.04
Leaf infection	133.52	6.01**	7.6
Stem infection	671.47	8.60**	14.2
Kernel infection	249.02	2.58**	15.8

**Significant at 1% level

DISCUSSION

Wide differences in reaction to Septoria were observed among the oat strains tested. As these differences were statistically significant and rather consistent from year to year, they can be considered as inherent. As was true in 1955, the most distinct strain differences were in degree of stem infection while the kernel infection was least distinct.

The reactions of the experimental selections indicate that Clinton, Vicland and other Victoria-Richland derivatives, and Hawkeye-Victoria derivatives frequently were one or more parents of the resistant oat selections. Andrew, Bond, Clarion, Rainbow, and Rodney frequently were one or more parents of the susceptible experimental selections. Other suggestions of variety contributions toward resistance or toward susceptibility are evident. Some crosses between resistant and susceptible varieties yielded both resistant and susceptible selections.

The Landhafer and Santa Fe varieties have been used recently as sources of resistance to crown rust (Puccinia coronata) and Hajira and Joanette varieties as additional sources of resistance to stem rust (Puccinia graminis avenae). Auxiliary tests indicate that these varieties are not highly susceptible to Septoria avenae and therefore it is unlikely that they have contributed to Septoria susceptibility of the experimental oat selections.

Many of the experimental oat selections with relatively low Septoria infections represent the best available combinations for desirable agronomic characteristics and resistance to other diseases. The data presented here should serve as a useful guide in choosing varieties for release or recommendation in regions where the Septoria disease is of major importance. These data would also appear to be valuable in the planning of future crossing programs.

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A NEW HELMINTHOSPORIUM DISEASE OF BERMUDA GRASS[1]

T. E. Freeman

Throughout the late summer and fall of 1956, Bermuda grass (Cynodon dactylon (L.) Pers.) in the Gainesville, Florida, area was found affected by a leaf-spot disease. Symptoms were atypical of those incited by the commonly occurring leaf-spot organism (Helminthosporium cynodontis Marig.); moreover, this apparently new disease was widespread throughout the entire area. These two facts prompted the initiation of studies to determine the nature of the disease.

SYMPTOMS

Symptoms of the disease consist of numerous small, elongated purplish-black spots on the leaves and sheaths (Fig. 1). These spots rarely exceed 2-3 in length and 0.5-1 mm in width, except in cases where they coalesce. In most instances the area immediately adjacent to the collar is

FIGURE 1. Bermuda grass leaves attacked by Helminthosporium stenospilum on left; healthy leaf on right (3X).

the most severely affected part of the leaf blade. This condition is probably due to the washing of spores to this area by dews and rain. Heavily spotted leaves turn yellow and in some cases are tinged with pink to reddish areas. Later, these leaves wither and assume a straw color. Helminthosporium cynodontis causes similar spots, but not so dark in color, being more reddish-purple and surrounded by a yellow halo during early stages. Haloes have not been noted in the case of the disease under study; instead, there is a sharp line of demarcation between the healthy green tissue and the diseased tissue.

CAUSAL ORGANISM

A species of Helminthosporium was found consistently fruiting on diseased material, especially if it had been maintained under moist conditions. Several isolations from diseased plants found in different areas yielded a high percentage of a species of Helminthosporium with essentially the same characteristics as the one found on fresh material maintained in a moist chamber. Bermuda grass was inoculated with a conidial suspension of the isolated fungus. After 48 to 72 hours in a moist chamber, symptoms typical of those found in the field were noted. The fungus could readily be isolated from the inoculated grass. These isolation and inoculation data indicate that the isolated fungus is the causal agent of the new leaf-spot disease.

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[1] Florida Agricultural Experiment Station Journal Series, No. 615.

Morphologically, the characteristics of the fungus resembled those of the brown stripe parasite of sugarcane (Helminthosporium stenospilum Drechs.). The spores were dark olivaceous, usually curved, 6-12 septate, and 62-118 x 12-17 microns. They were thick-walled, with the wall on the convex side thicker than that on the concave side, a characteristic noted by Faris (2) for H. stenospilum. To further verify the identity of the fungus, inoculations were made on two stripe-susceptible sugarcane varieties. Typical brown stripe symptoms (2, 3) appeared on both varieties as a result of these inoculations. Therefore, it appears that H. stenospilum [2] is the causal agent of the Bermuda grass disease studied. To the knowledge of the author, this is the first report of this fungus causing a disease of Bermuda grass.

FURTHER STUDIES

Since the disease was apparently unreported before, further studies concerning it were warranted. Therefore, tests were designed to compare the severity of the new disease with that of the more commonly recognized one incited by Helminthosporium cynodontis. These tests were also designed to study the reaction of different varieties and selections of Bermuda grass to both diseases. Studies were initiated, using a factorial design that included seven varieties and three of Dr. G. C. Nutter's selections,and the two pathogens. The experiment was carried out under greenhouse conditions, using potted plants. Each of the 20 treatment combinations was replicated 4 times. Comparable conidial suspensions of the two fungi were used for inoculation and each replication was subjected to approximately the same amount of inoculum. Five days after inoculation, a comparative disease severity rating was applied to each replication of each treatment. The ratings ranged from 1 to 5 with 5 being the most severe. The results of the test are shown in Table 1.

Table 1. Comparative disease severity ratings[a] of two species of Helminthosporium on various Bermuda grasses.

Variety or selection	Severity rating		
	H. cynodontis	H. stenospilum	Mean
Everglades	2.3	5.0	3.65
Bayshore	2.8	4.8	3.80
Ormond	2.5	3.8	3.15
Fla. 49	1.5	5.0	3.25
Fla. 50	3.3	4.8	4.05
Fla. 51	2.5	5.0	3.75
Tiflawn	2.8	4.5	3.65
Tiffine	2.0	3.8	2.90
Tifgreen	1.3	3.3	2.30
Common	2.8	4.3	3.55
Mean	2.38	4.43**	

LSD for varieties: .01 level .79; .05 level .59
LSD for interaction: .01 level no significance; .05 level .98
** Significant at the .01 level.

[a]Average of four replications.

The data presented in Table 1 show that in all cases H. stenospilum caused significantly more damage to the varieties and selections tested than did H. cynodontis. None of the materials tested showed complete resistance to either species. Observations on the prevalence of H. stenospilum on Bermuda grass in the State are not complete. The parasite has been known to occur in the State since 1928 (1); therefore, it is likely that it is widespread in the

[2]Thanks are extended to Dr. E. S. Luttrell for his aid in identification of the fungus.

area. This being the case, the data in Table 1 indicate that H. stenospilum may be found to be a more important pathogen on Bermuda grass in Florida than the commonly recognized H. cynodontis.

Literature Cited

1. DRECHSLER, C. A. 1928. A species of Helminthosporium distinct from Helminthosporium sacchari, causing brown stripe of sugar cane. (Abst.) Phytopathology 18: 135-136.
2. EDGERTON, C. W. 1955. Sugarcane and its Diseases. Louisiana State University Press, Baton Rouge, Louisiana, pp. 101-102.
3. FARIS, J. A. 1928. Three Helminthosporium diseases of sugar cane. Phytopathology 18: 753-774.

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BARLEY STRIPE MOSAIC IN WINTER WHEAT

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INTRODUCTION

Barley stripe mosaic has been recognized as a serious disease of barley. The nature of the disease suggests that it may be a potentially serious threat to winter wheat.

McKinney (4) has shown that the disease is caused by a seedborne virus. This work has been confirmed by Eslick (1). McKinney (5) has also shown that the barley stripe mosaic virus is readily transferred from diseased plants to healthy plants by contact of plant parts caused by air movements or handling. He has also shown that this virus can be transferred by manual inoculation to wheat, sweet corn, and some of the grasses. It seems to be possible, therefore, that barley stripe mosaic virus might be transferred from diseased barley to healthy wheat when the two crops are grown adjacent to each other or when volunteer barley plants are present in wheat fields. The spread of the disease by cross-pollination has been suggested by the presence of rod-shaped particles in seed produced from healthy pistils when pollinated by pollen from diseased plants. The association of the rod-shaped particles and the virus disease has been established by Gold, et al. (2).

EXPERIMENTAL PROCEDURE

Four varieties of winter wheat, Wasatch, Cache, Columbia, and C. I. 12932, were seeded in two-row plots 12 feet long in the disease nursery at the Aberdeen Branch Experiment Station, Aberdeen, Idaho, on October 3, 1955. All rows of wheat were spaced 2 feet apart. On April 1, 1956, each row of wheat was flanked by a single row of stripe mosaic virus infected barley sown 6 inches from the wheat. Infected seed of the barley varieties, Trebi and Glacier, was used. Infection in the barley seed ranged from 25 to 50 percent. On May 1 and May 14 the wheat varieties were clipped to a height of 6 inches to allow the spring barley varieties to become established. Air movements produced a rubbing action between the barley and wheat leaves throughout the growing season. Each row of wheat was harvested individually in August. Seed samples from the eight treatments were assayed for the virus in the greenhouse at North Dakota State College, Fargo, North Dakota. The results are shown in Table 1.

Table 1. Percent infection of barley stripe mosaic in winter wheat grown adjacent to diseased barley.[a]

Wheat variety	Grown with diseased barley variety	No. plants tested	Number with symptoms	Percent infection
Wasatch	Trebi	125	4	3.2
	Glacier	147	3	2.0
Cache	Trebi	110	1	tr[b]
	Glacier	205	3	1.5
Columbia	Trebi	319	1	tr
	Glacier	238	1	tr
C.I. 12932	Trebi	147	4	2.7
	Glacier	105	0	0.0

a The wheat varieties were grown from seed lots which failed to produce any evidence of the virus in a 1955 greenhouse assay.

b tr - denotes trace.

[1]Agronomists, and Plant Pathologist, respectively, Crops Research Division, Agricultural Research Service, United States Department of Agriculture, Aberdeen, Idaho, and Fargo, North Dakota, in cooperation with the Idaho and North Dakota Agricultural Experiment Stations.

DISCUSSION AND CONCLUSIONS

The data presented indicate that barley stripe mosaic can be transferred to winter wheat under natural conditions. Data are not yet available as to the buildup of the virus in a population of wheat plants sown with diseased seed. It is not known at what rate the virus will be transferred from diseased to healthy wheat. However, if the disease spreads in wheat as rapidly as it does in barley, there is cause for alarm. This is especially true in view of the yield reductions reported by McNeal, Berg, Afanasiev, and Army (6) for spring wheat and by Eslick (1) for barley. Hagborg (3) has found in both field and greenhouse tests that large and significant reductions in height, yield, kernel weight, and grade of the crop resulted from early inoculations with the barley stripe mosaic virus. Yield reductions were approximately 75 percent for wheat and 64 percent for barley.

Limited data from early work on barley stripe mosaic indicate that particular attention should be given this disease in wheat. Assuming that symptom expression in wheat is dependent upon environmental conditions, degree of infection, and the time of infection, as has been reported for barley (5), critical observations will be necessary to detect the presence of the virus in wheat before it has become widespread.

Literature Cited

1. ESLICK, R. F. 1953. Yield reductions in Glacier barley associated with a virus infection. Plant Dis. Reptr. 37: 290-291.
2. GOLD, A. HERBERT, COIT A. SUNESON, BYRON R. HOUSTON, and JOHN W. OSWALD. 1954. Electron microscopy and seed and pollen transmission of rod-shaped particles associated with the false stripe virus disease of barley. Phytopathology 44: 115-117.
3. HAGBORG, W. A. F. 1954. Dwarfing of wheat and barley by the barley stripe mosaic (false stripe) virus. Canad. Jour. Bot. 32: 24-37.
4. McKINNEY, H. H. 1951. A seed-borne virus causing false-stripe symptoms in barley. Plant Dis. Reptr. 35: 48.
5. McKINNEY, H. H. 1954. Cultural methods for detecting seed-borne virus in Glacier barley seedlings. Plant Dis. Reptr. 38: 152-162.
6. McNEAL, F. H., M. A. BERG, M. M. AFANASIEV, and T. J. ARMY. 1957. The influence of barley stripe mosaic on yield and other plant characters of 8 spring wheat varieties grown at 4 nitrogen levels. Agron. Jour. (in press).

CROPS RESEARCH DIVISION, AGRICULTURAL RESEARCH SERVICE, IN COOPERATION WITH THE IDAHO AND NORTH DAKOTA AGRICULTURAL EXPERIMENT STATIONS

TRISTEZA AND SEEDLING YELLOWS OF CITRUS

James M. Wallace [1]

Abstract

The seedling yellows reaction in lemons, described in New South Wales by Fraser, and in South Africa by McClean and van der Plank, is caused by a virus found commonly in those countries in association with what has been considered to be typical or ordinary tristeza. In California, seedling yellows virus has not been encountered in sweet orange or other citrus known to have been naturally infected with tristeza (quick decline) virus. Recent studies show that the causal virus of seedling yellows is carried by plants of Meyer lemon, Satsuma orange, and other citrus varieties known to be infected with tristeza virus and believed to have been carriers of the virus when orginally imported to the United States. Work is in progress to determine if the seedling yellows reaction is caused by a virus distinct from tristeza virus, as suggested by Fraser, or if it is a strain of the latter.

In 1952, Fraser (1) described a seedling yellows reaction on Eureka lemon, grapefruit, and sour orange and demonstrated that the causal virus was present in a very high percentage of the sweet orange and mandarin trees in New South Wales, but it was not found occurring naturally in trees of grapefruit, sour orange, or lemon. From these latter species, Fraser commonly obtained virus that she considered to be the tristeza-stem pitting complex. Using seedlings of West Indian lime and Eureka lemon as test plants, it was shown that in nature, seedling yellows was always accompanied by "tristeza-stem pitting complex." On the other hand, as mentioned above, only the tristeza virus was found in naturally infected grapefruit, sour orange, and lemon trees. Fraser concluded that the severe reaction on Eureka lemon seedlings (seedling yellows)"...is caused by a virus distinct from the tristeza-stem pitting complex."

McClean and van der Plank (2) in South Africa studied the seedling yellows reaction in relation to tristeza. In general, these workers agreed with Fraser in regard to symptomatology, host range, and the universal presence of the "stem pitting component" in mixture with the "seedling yellows component." However, McClean and van der Plank have concluded that tristeza is caused by a virus complex and that seedling yellows is a part of that complex.

In California, the early studies on tristeza (quick decline) virus dealt largely with the virus occurring naturally in sweet orange. Inoculations of seedlings of Mexican lime from these infection sources have given evidence of the existence of different strains of the virus, but inoculations to sour orange, Eureka lemon, and grapefruit seedlings gave no symptoms. Thus it seemed that seedling yellows, as described in New South Wales and South Africa, was not present in California in sweet orange trees which had become infected with tristeza by means of insect transmission of the virus.

It has been reported that Meyer lemon, numerous Satsuma orange varieties, and other kinds of citrus are believed to have been infected with tristeza when introduced into the United States (3). A study was begun recently to determine if the virus carried by these varieties differs from the virus of tristeza present in naturally infected sweet orange trees in California. Eureka lemon seedlings were inoculated from a selected group of trees of different species and varieties which had been shown to be infected with tristeza by earlier indexing to Mexican lime. The results of the first inoculations made in this connection indicate that the seedling yellows virus is present in 8 of 14 sources tested.

The experimental procedure was as follows: Three Eureka lemon seedlings, about 10 inches tall, were inoculated from 14 sources by means of small twig grafts. Twelve of these sources were trees of miscellaneous citrus in the variety collection of the Citrus Experiment Station, all of which are belived to have been infected with the virus of tristeza when they, or their parent types, were originally received at Riverside. One inoculum source was a tree of

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Table 1. Results of tests for seedling yellows virus in citrus clones suspected of having been infected with tristeza virus when introduced to California.

Source of virus	Origin & date received at Riverside[a]	No. of lemon sdlgs. in test	Reaction of lemon seedlings[b]		
			0	1	2
Kawano Wase Satsuma	Japan - 1928	3			3
Meyer lemon, C.E.S. 1-54	China - 1917	3			3
Satsuma orange, C.E.S. 266	Florida - 1914	3		3	
Waialua orange	Hawaii - 1914	3			3
Red Ling Mung lemon, U.C.L.A.	China - 1931	3			3
Parsons Special mandarin	Florida - 1914	3		1	2
Ponds Seedless lime	Hawaii - 1914	3		3	
Hawaiian shaddock	Hawaii - 1914	3	1	2	
Batangas mandarin	Phil. Is. - 1914	3			3
Ponkan mandarin, C.E.S. 2593	China ? - 1937	3		1	2
Ponkan mandarin, C.E.S. 2329	China ? - 1930	3		3	
South African lemon	So. Africa - 1917	3			3
Cecily grapefruit	So. Africa - 1930	3	1	2	
Valencia orange[c] (naturally infected orchard)	California	3		3	
Non-inoculated controls		6	6		

[a]Plants or budwood received directly from place indicated or through United States Dept. of Agriculture, Office of Foreign Plant Introduction.

[b]0 = No reaction; 1 = Mild reaction, slight stunting only; 2 = Seedling yellows reaction, stunting and yellowing.

[c]Mexican lime seedlings infected from this source by Aphis gossypii supplied inoculum for lemon seedlings.

Red Ling Mung lemon located at the University of California at Los Angeles. As a check on these, Eureka seedlings were inoculated with tristeza virus obtained from a naturally infected Valencia orange tree in a commercial grove. Six Eureka seedlings which received no inoculation were included for comparison with the inoculated seedlings. At the time the inoculation grafts were made the seedlings were decapitated a short distance above the grafts.

One month after treatment, the new growth on seedlings of several groups clearly showed evidence of seedling yellows. As time progressed these plants displayed small, chlorotic leaves and very slow growth. In some instances some of the leaves dropped and in later stages there was very little stem elongation and leaf formation. The lemon seedlings inoculated from the naturally infected Valencia orange and from 5 other sources did not develop symptoms of seedling yellows. Nearly all of the seedlings of these 6 groups, however, were slightly stunted as compared to non-inoculated seedlings. Figure 1 shows the two symptom reactions in comparison with a virus-free plant.

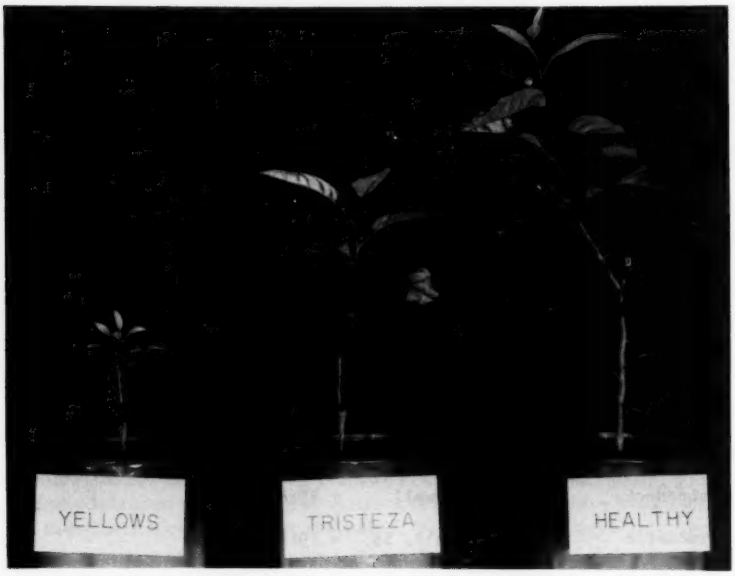

FIGURE 1. Seedling yellows and tristeza reactions on Eureka lemon seedlings. Left, Inoculation from tristeza-infected South African lemon (import). Center, Inoculation from naturally-infected Valencia orange. Right, Non-inoculated check. Photographed 4 months after inoculations.

Detailed results of this test are presented in Table 1. The numbers of plants showing no reaction, mild reaction, and seedling yellows are recorded. In two instances, one plant of a group showed the mild reaction and the remaining two plants had seedling yellows. Tests have not been completed to determine if this represents a separation of a mild component of the virus complex.

These preliminary studies demonstrated that the virus of seedling yellows is present in some tristeza-infected citrus in California. Other work is in progress to study further the relationship of seedling yellows virus to the tristeza virus strain or complex commonly found in naturally infected citrus trees. If it can be demonstrated that the virus of seedling yellows is not present in naturally infected trees in California, this will suggest either that the insect vector (Aphis gossypii Glover) is incapable of transmitting seedling yellows virus, or that the original sources of infection in California citrus plantings were free of seedling yellows and thus only the "tristeza-stem pitting" virus referred to both by Fraser (1) and by McClean and van der Plank (2) has so far become distributed in citrus here.

Presently, the writer favors the opinion of McClean and van der Plank that the virus that causes seedling yellows is a part of the tristeza complex and not a distinct virus. Studies in progress include cross protection reactions, insect vector relationships, and efforts to obtain the seedling yellows strain of virus separate from the ordinary tristeza virus. The discovery of seedling yellows in California will also provide an opportunity to make other investigations which could explain some of the differences or lack of agreement in symptomatology, host reactions, and vector relationships, etc., reported by various workers in different countries where tristeza has been investigated.

The probability exists, it seems, that the seedling yellows reaction is merely a strain of tristeza virus which causes the reaction on lemon seedlings and in addition produces a reaction on lime seedlings identical to that resulting from infection with what Fraser described as the "tristeza-stem pitting complex." It may be possible to explain the situation by means of studies on insect vector relationships and host range of the two viruses or virus strains.

Literature Cited

1. FRASER, LILIAN. 1952. Seedling yellows, an unreported virus disease of citrus. Agr. Gaz. N. S. Wales 63: 125-131.
2. McCLEAN, A. P. D., and J. E. VAN DER PLANK. 1955. The role of seedling yellows and stem pitting in tristeza of citrus. Phytopathology 45: 222-224.
3. WALLACE, J. M., P. C. J. OBERHOLZER, and J. D. J. HOFMEYER. 1956. Distribution of viruses of tristeza and other diseases of citrus in propagative material. Plant Dis. Reptr. 40: 3-10.

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SUMMER DWARF OF STRAWBERRY[1]

D. A. Slack[2], Carter Seymour[3], Henry Fields[2] and J. P. Fulton[2]

Summary

Recent occurrences of summer dwarf (Aphelenchoides besseyi) were recorded in strawberry plantings. Aberrant symptoms of the disease were observed on plants in the field and greenhouse. Field development was most striking during late fall rather than mid-summer. The problem is apparently general rather than local since diseased plants originated in Arkansas, Tennessee, Oklahoma, Missouri and Maryland.

Strawberry dwarf or crimp is a disease caused by a foliar or bud nematode belonging to the genus Aphelenchoides Fisher. Attention was focused on this disease as it occurred in the United States following a report by Plakidas (10) from Louisiana in 1928. Subsequent work reported by Christie (2, 3, 5), Christie and Boyd (6), and Brooks (1) established the disease syndrome. It remained for Christie (3, 4) and Christie and Crossman (7) to differentiate between spring dwarf, caused by A. fragarie (Ritzema Bos) Christie, as it occurred in Massachusetts, and summer dwarf, caused by A. besseyi Christie, the disease associated with plants grown in more southern areas.

During the 1930's summer dwarf was found to be common in strawberry-growing districts of the Southern States and constituted a definite burden on the strawberry industry (1, 5, 9). Since 1942, summer dwarf has received little attention. Growers producing plants for sale and inspectors responsible for inspection of plants were cognizant of the problem. It is assumed that strawberry fields were eliminated as sources of plants where summer dwarf was present, and that the disease was correspondingly reduced in importance.

DEVELOPMENT IN ARKANSAS

In 1954 a disease problem of a questionable nature was observed in a few fields of strawberries in Arkansas. As indicated later, the disease proved to be summer dwarf. The disease was found in plantings set from inspected plants of the Blakemore variety and, in many instances, on land not planted to strawberries previously. The disease was not observed in the field until August. Symptom expression was most pronounced after September 15 and continued into December. After December 15, plants reverted to a quasi dormant condition and symptoms were difficult to observe. In 1955, plants that exhibited the disease the previous year appeared to have recovered from the debility. There was a reduction in yield from some diseased areas and an increase in amount of misshapen fruit. In August, however, the disease reappeared and had spread to encompass a larger number of plants in a roughly ovoid area in the affected fields. Specimens of the diseased plants were removed from the field. One lot was planted in the greenhouse and a second lot taken into the laboratory for observation. Of the plants taken into the laboratory, all yielded specimens of Aphelenchoides besseyi. Diseased plants transferred to the greenhouse developed symptoms typical of summer dwarf. Leaf petioles of new growth were short and thickened, and exhibited a reddish cast. Leaflets were narrow in proportion to length and severely distorted and crinkled. In addition, phyllody of flowers produced by some diseased plants was noted. Fruit borne by diseased plants was malformed (Fig. 1, 2, and 3). Symptoms similar to those described were produced in strawberry plants artificially inoculated with A. besseyi recovered from buds of diseased plants. In some respects diseased plants resembled a condition attributed to aster yellows (8, 11). The disease was not lethal and diseased plants lacked severe chlorosis and development of adventitious crowns.

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[1]Published with approval of the Director of the Arkansas Agricultural Experiment Station, Fayetteville, Arkansas, and Chief Inspector of the State Plant Board, Little Rock, Arkansas.

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FIGURE 1. Malformed fruit and distorted leaves typical of strawberry plants with summer dwarf.

FIGURE 2. Phyllody of a flower occasionally associated with summer dwarf.

FIGURE 3. Later development of phyllody showing partial fruit development.

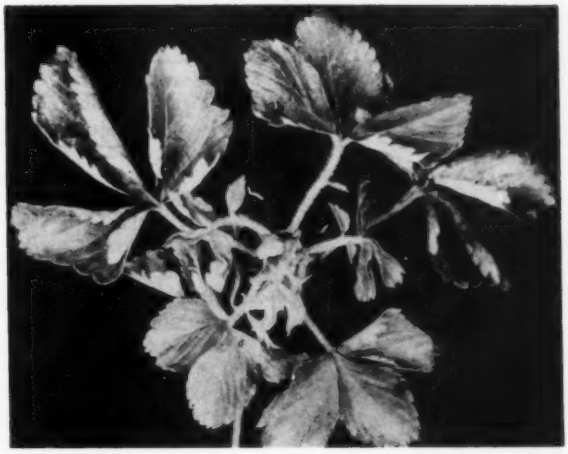

FIGURE 4. Symptoms of summer dwarf on field-grown plant collected in late October. The upward curling of leaflets and the chlorotic, dwarfed and twisted new leaves are shown.

In the field, plants diseased with summer dwarf were characterized by outspread older leaves frequently showing an upward curling of leaflets. New leaves were dwarfed, chlorotic and somewhat twisted (Fig. 4). A slight crimping effect was apparent on a few younger leaflets; however, malformation of leaflets was slight as compared with that described by Christie (5) and Brooks (1). Summer dwarf was observed most commonly in plants of the Blakemore variety. Approximately 87 percent of the acreage devoted to strawberry plant production was planted to Blakemore.

PROCEDURE USED IN DIAGNOSING SUMMER DWARF

In an effort to eliminate dwarf from strawberry fields grown for plants, inspectors for the State Plant Board were briefed on a revised inspection procedure. Date of inspection was set from September 15 through November. Fields were checked for plants exhibiting possible symptoms of summer dwarf. In questionable fields, a minimum sample of 50 plants per acre, taken from at least 15 locations at the inspector's discretion, were dug and sent to the laboratory to determine the presence or absence of A. besseyi in the buds. In the laboratory the bud was removed from each plant, macerated separately in a Syracuse watch glass and covered with distilled water. Approximately 1 to 2 hours after maceration, each bud was checked using a stereoscopic microscope at 30X magnification. Diseased plants were not heavily infested with A. besseyi; each bud usually yielded from 10 to 30 specimens. Nematodes recovered were checked periodically for species determination. Based on laboratory diagnosis, strawberry fields were quarantined or released for plant production.

SOURCE OF INFECTED PLANTS

An attempt was made to correlate source of plants with occurrence of summer dwarf. The history of the plants set in each diseased field during 1955-56 and 1956-57 was acquired for the previous seasons. Information was vague in some instances as to source of plants, since an individual may handle plants from contract growers, or resale plants that had passed two field inspections grown in other areas especially Tennessee. From the 1951-52 through the 1955-56 growing seasons the quantity of Arkansas-grown strawberry plants available to growers was reduced by 68 percent of the 10-year average from 1930-1940 by drought and heat. Plants were purchased from various sources to offset the reduced volume, and to introduce new varieties and plants labeled "virus-free" into the plant production program.

Table 1. Source of strawberry plants set in fields that developed summer dwarf.

Fields with summer dwarf [a]		Source of plant shipment (Year)		
No.	Year	1955-56	1954-55	1953-54
11	1955-56		Arkansas	Tennessee
6	1955-56		Arkansas	Arkansas-Tennessee
2	1955-56		Arkansas	Maryland
3	1955-56		Arkansas	Arkansas
1	1955-56		Missouri	Tennessee
2	1955-56		Arkansas	Oklahoma
3	1956-57	Tennessee		
1	1956-57	Arkansas		

[a] 126 acres were quarantined due to summer crimp in 1955-56; 11 acres were quarantined in 1956-57.

As indicated in Table 1 plants that exhibited summer dwarf during the fall of 1955-56 primarily originated from fields in Arkansas the previous year. Plants in many of these fields exhibited summer dwarf during 1954-55; however, the disease was not recognized as such and recorded. It would appear that plants diseased with summer dwarf originated from fields in Arkansas, Tennessee, Oklahoma, Missouri and Maryland and were transferred to growers in shipments during 1953-54. During the fall of 1956, plants in four strawberry fields exhibited excessive summer dwarf; three fields were set with plants from Tennessee and one was set with plants from Arkansas.

DISCUSSION

Since 1940, summer dwarf has been of little importance in strawberry-growing areas of the South. Lack of attention in plant production could allow this disease to assume a more serious status. Several factors probably influenced the expression of summer dwarf of strawberry during recent years: 1) Populations of Aphelenchoides besseyi recovered from diseased strawberry buds were relatively low. 2) Relatively dry, hot weather during mid-season retarded plant growth and disease development. 3) Flush of plant growth in the fall was accompanied by disease development. Due to the aberrant behavior of summer dwarf, field diagnosis alone is sometimes difficult or impossible. Since the laboratory procedure involved in demonstrating the presence of A. besseyi in buds of diseased plants is not cumbersome or time consuming and does not require excessive equipment, laboratory analysis could supplement field diagnosis.

With the increased emphasis on development and planting of "virus-free" strawberry varieties, other disease problems such as summer dwarf are easily overlooked or ignored by plant producers and inspection officials. The opportunity to develop a disease-free strawberry program as opposed to a "virus-free" program would appear to have merit.

Literature Cited

1. BROOKS. A. N. 1931. Crimp -- a nematode disease of strawberry. Univ. of Florida Agr. Exp. Sta. Bul. 235. 27 pp.
2. CHRISTIE, J. R. 1932. Recent observations on the strawberry dwarf nematodes in Massachusetts. Plant Dis. Reptr. 16: 113-114.
3. CHRISTIE, J. R. 1938. Two distinct strains of the nematode Aphelenchoides fragarie occurring on strawberry plants in the United States. Jour. Agr. Res. 57: 73-80.
4. CHRISTIE, J. R. 1942. A description of Aphelenchoides besseyi, n. sp., the summer dwarf nematode of strawberries, with comments on the identity of Aphelenchoides subtenuis (Cobb, 1926) and Aphelenchoides hodsoni Goodey, 1935. Proc. Helm. Soc. of Wash. 9: 82-84.
5. CHRISTIE, J. R. 1943. Spring dwarf and summer dwarf of strawberries. U. S. Dept. Agr. Circular 681. 10 pp.
6. CHRISTIE, J. R., and O. C. BOYD. 1934. Aphelenchoides fragariae on Cape Cod strawberries. Plant Dis. Reptr. 18: 45-46.
7. CHRISTIE, J. R., and LOUISE CROSSMAN. 1936. Notes on the strawberry strains of the bud and leaf nematode Aphelenchoides fragarie. Proc. Helm. Soc. of Wash. 3: 69-72.
8. FRAZIER, N. W., and H. E. THOMAS. 1953. Strawberry a host of western aster yellows virus. Plant Dis. Reptr. 37: 272-275.
9. MOOK, P. V. 1931. Survey for dwarf disease of strawberries. Plant Dis. Reptr. 31: 14-15.
10. PLAKIDAS, A. G. 1928. Strawberry dwarf. Phytopathology 18: 439-444.
11. PLAKIDAS, A. G. 1951. Chlorotic phyllody of strawberry. Plant Dis. Reptr. 35: 495-496.

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NEMATODE GENERA FOUND IN NEW YORK STATE ORCHARDS

W. F. Mai[1], James Dolliver[2], J. D. Kirkpatrick[3], and K. G. Parker[1]

During the 1956 growing season a survey was made of the potentially pathogenic nematodes present in and around the roots of tree fruits growing in New York State. Samples were collected from 305 sour cherry trees in 81 orchards, 38 apple trees in 10 orchards, and 5 peach trees in 2 orchards (Table 1). Seventeen of the sour cherry orchards were included in the survey because the growth pattern of the trees indicated a nematode problem (2). Sixty-four additional cherry orchards were sampled in order to determine the kinds of potentially parasitic nematodes present in orchards varying with respect to vigor, age, location, soil type, and cultural practices. The apple orchards examined all exhibited poor growth unexplained on the basis of soil and cultural conditions.

PROCEDURE

In taking the soil samples, after removing the top few inches of soil, plant growth, or mulch, a hole 18 inches deep was dug with a post hole digger. Soil for the sample was obtained from the sides of the hole. All tree roots present in the soil removed by the post hole digger were collected. Sometimes small roots on the trees were very sparse and it was necessary either to dig a larger area of soil or move to a different tree to obtain an adequate root sample. Usually this was the result of death of the rootlets because of nematode attack (2).

For the determination of endoparasitic nematodes, the roots were incubated according to the method described by Young (3). The soil samples were processed by the Christie and Perry technique (1). To determine whether individuals of the genus Criconemoides -- which do not move readily through the tissue paper of the Baerman funnel -- were present, a sample of material from the 325- or 270-mesh screen was examined under the microscope, directly after removal from the screen or after centrifugation.

RESULTS

Potentially pathogenic nematodes were found in each of the orchards examined (Table 1). The order of frequency of occurrence of the several genera was approximately the same in the different tree fruit orchards and in each of the two categories of sour cherry orchards. Pratylenchus was the genus encountered most frequently. The frequency of occurrence of Pratylenchus was essentially the same whether the results of soil samples only were taken into account or whether both root and soil samples were considered, as shown in Table 1.

The wide distribution of these genera indicates that the occurrence of a potentially pathogenic species does not necessarily demonstrate the presence of a nematode problem. In fact, Pratylenchus penetrans, a species shown by greenhouse and field tests to cause growth reduction, may occur in orchards in which the trees, as judged by usual standards, are in good vigor. Furthermore, the numbers of P. penetrans found cannot be relied upon for the determination of a nematode problem. The feeder roots may be so severely damaged by the nematodes that the population of the nematodes themselves is greatly reduced.

To summarize, it appears that potentially pathogenic nematodes are distributed widely throughout the orchard soils of New York State. In some orchards the evidence is strong that attack by Pratylenchus penetrans is an important factor in the decline of cherry and to a lesser extent of apple trees.

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Table 1. Nematode genera found in New York State orchards.

Nematode genus	Orchards			
	No. sampled	No. infested	No. sampled	No. infested
A. Sour cherry orchards selected to include examples of a wide variety of orchard types.				
Pratylenchus sp.[a]	64	64	255	247
Xiphinema sp.	64	59	255	176
Paratylenchus sp.	64	41	255	66
Tylenchus sp.	64	37	255	71
Criconemoides sp.	64	31	255	68
Tylenchorhynchus sp.	64	11	255	15
Ditylenchus sp.	64	2	255	2
Hemicycliophora sp.	64	3	255	3
B. Sour cherry orchards with an apparent nematode problem.				
Pratylenchus sp.[a]	17	17	50	49
Xiphinema sp.	17	11	50	26
Paratylenchus sp.	17	8	50	24
Criconemoides sp.	17	5	50	9
Tylenchus sp.	17	4	50	9
Tylenchorhynchus sp.	17	2	50	3
C. Apple orchards with an apparent nematode problem.				
Pratylenchus sp.[a]	10	8	38	36
Xiphinema sp.	10	4	38	5
Tylenchus sp.	10	3	38	4
Paratylenchus sp.	10	2	38	4

Table 1 (con't.)

D. Peach orchards selected at random.				
Pratylenchus sp.[a]	2	2	5	5
Xiphinema sp.	2	1	5	1
Paratylenchus sp.	2	1	5	1
Criconemoides sp.	2	1	5	2
Totals (all orchards)	93	91	348	337

[a] All individuals that were examined under a compound microscope were identified as Pratylenchus penetrans.

Literature Cited

1. CHRISTIE, J. R., and V. G. PERRY. 1951. Removing nematodes from soil. Proc. Helm. Soc. Wash. 18: 106-108.
2. PARKER, K. G., and W. F. MAI. 1956. Damage to tree fruits in New York by root lesion nematodes. Plant Dis. Reptr. 40: 694-699.
3. YOUNG, T. W. 1954. An incubation method for collecting migratory endoparasitic nematodes. Plant Dis. Reptr. 38: 794-795.

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CONTROL OF THE CITRUS NEMATODE AND PHYTOPHTHORA SPP. BY VAPAM

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Young citrus trees usually grow poorly when attacked by the citrus nematode Tylenchulus semipenetrans, or by Phytophthora spp. (brown-rot fungi) and other soil fungi. These pests frequently occur in soils that are being replanted to citrus, and compounds that possess both fungicidal and nematocidal properties are desirable for their control.

Vapam (sodium n-methyl dithiocarbamate) is water-soluble and possesses both fungicidal and nematocidal properties. In the presence of water and surface active materials, such as soil, Vapam changes rapidly to methyl isothiocyanate and other compounds. Methyl isothiocyanate is volatile and only slightly soluble in water.

Vapam applied in water in basins (tests reported below) usually effectively controlled the citrus nematode and Phytophthora citrophthora and P. parasitica. The application of Vapam in water in basins, however, has certain limitations, especially when large areas or sloping lands are being treated. Thus, other methods of application might be more desirable. This report deals with the efficacy of Vapam applied by seven methods in the field and suggestions for its use in treating old citrus soils prior to replanting. In these tests a special effort was made to control the pathogenic organisms to depths of four or more feet, since citrus trees usually are long lived and likely would soon become infected otherwise. The data presented are pertinent to control of the citrus nematode and Phytophthora spp. Chemicals applied to kill parasitic organisms usually also kill nonparasitic types. Thus, a relative measure of the fungicidal effects of a chemical treatment may be obtained by determining the effects on the fungus population of the soil. This was done in connection with several of the tests described below.

METHODS

All of the tests, except one, were made in the field on sandy loam, fine sandy loam, and loam soils. The soils were undisturbed below the depth of tillage (8-12 inches). For many of the basin treatments, metal rings 44 inches in diameter and 12 inches high were driven into the soil so that they retained water (Fig. 1). Forty gallons of water in these basins was equivalent to water 6 inches deep.

When Vapam was applied in irrigation water in furrows or by sprinklers the desired amount of Vapam was metered uniformly into the intake of a 2-inch centrifugal pump over a 12- to 27-hour period (Fig. 2). The metering device for the Vapam consisted of a small power sprayer or carbon dioxide pressurized containers and small orfices through which the Vapam was delivered into the water.

The furrow applications were made on plots 24 feet wide and 400 feet long. The furrows were 20 inches apart and ran the long axis of the plots as shown in Figure 3. Sufficient solution was introduced at the head end of the furrows so that there was a slight run-off at the end of the plots.

One-inch plastic hoses fitted with low head revolving sprinklers were used for the sprinkler treatments. The hoses were attached to a centrifugal pump and sufficient pressure was maintained so that the sprinklers delivered approximately 0.4 inches of solution over the surface of the plots each hour.

A conventional tractor-drawn shank type of equipment was used for applying the Vapam in the soil at depths of 5 to 7 inches. In all of the treatments the shanks were 12 inches apart. After applying the Vapam the surface was cultipacked or rolled and then furrows were made

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[2]The authors gratefully acknowledge the assistance of the following in many of the tests: L. J. Klotz, J. O. Erwin, and T. E. Szuszkiewicz, members of the University of California Agricultural Experiment Station; and of Marvin Miller and C. D. Gustafson, Farm Advisors, University of California Agricultural Extension Service; and F. J. Foote, Director of Research, Limoneira Company, Santa Paula, California. Growers mentioned in the text made available facilities on their properties and also cooperated in other ways in the tests.

FIGURE 1. Movable metal basins used in testing the nematocidal and phytotoxic properties of Vapam in an orange grove. Different concentrations of Vapam in water are being applied.

FIGURE 2. Equipment used to apply Vapam in water to field plots. The Vapam concentrate in two containers "A" is metered through a small orifice "B" into the source of water for the centrifugal pump "C".

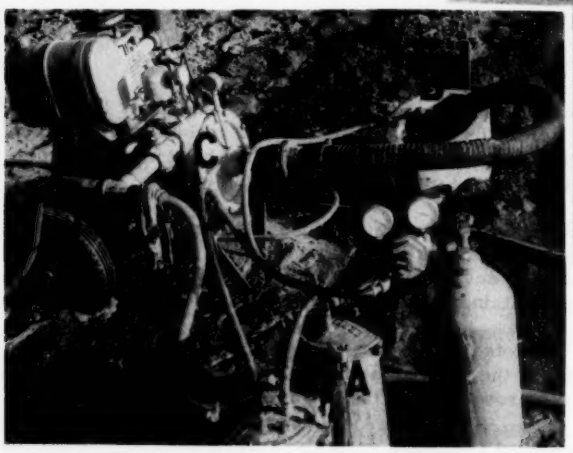

FIGURE 3. Vapam being applied in water by furrows and by sprinklers on adjacent plots at the Limoneira Ranch, near Santa Paula. A plot treated by injecting Vapam into the soil is in the lower right-hand corner.
Photo by R. G. Platt

20 inches apart for irrigation. However, when irrigated by sprinklers the surface was not cultipacked.

Soil samples for determination of the citrus nematode were taken with a 1-inch soil tube. Four soil cores per plot were composited for a sample. The number of live citrus nematodes in 50 milliliters of soil was determined by a modification of the Baermann funnel method.

The Phytophthora species were determined by placing soil in pans, adding water, and then placing surface-sterilized lemons on the saturated soil. The fungi were isolated from any decay which developed, and identified. Other soil fungi were determined by the dilution plate method, using peptone, dextrose, and rose bengal-streptomycin media[3].

The mechanical analysis of two of the soils was made by conventional methods and is presented in Tables 1 and 2. Both soils were alluvial types and showed stratification. Wet soil paste was used for the pH determinations.

EXPERIMENTS AND RESULTS

Vapam Applied in Basins on a Sandy Loam Soil: -- The effectiveness of two concentrations of Vapam in water and of different amounts of the solution for control of the citrus nematode were tested at Riverside in 1955. Forty and 80 gallons of water containing 50 and 100 ppm of Vapam were placed in the metal basins described above. These were equivalent to 6- and 12-inch depths of the Vapam solutions and to 68.5, 137, and 274 pounds of active ingredient per acre[4].

Approximately 92 percent of the citrus nematodes in the first foot, and 45 and 46 percent in the second and third foot of soil were killed when 6 surface inches of solution containing 50 ppm of Vapam (68.5 pounds per acre) were applied (Table 3). When either 12 surface inches of 50 ppm Vapam solution or 6 inches of 100 ppm Vapam (137 pounds per acre) were applied, control was slightly better, but unsatisfactory. However, 12 surface inches of solution containing 100 ppm of Vapam (274 pounds per acre) killed 100 percent of the citrus nematodes to a depth of 36 inches and 85 percent in the fourth foot.

Vapam at the lowest concentrations used, 6 surface inches of water containing 50 ppm, killed the orange feeder roots in the top 2 feet of soil and did not control the citrus nematode.

Approximately 1 inch of water was required to wet 1 foot of the soil to field capacity. Thus, both the 6 and the 12 surface inch treatments of Vapam solutions wet the soil to a depth greater than depths at which the citrus nematode was killed. It appears that some of the Vapam was sorbed on the soil, and the concentration in the water thereby decreased as it moved through the soil.

Vapam Applied in Basins on a Loam Soil:— The effectiveness of Vapam for control of the citrus nematode on a loam soil, which contained 22 to 32 percent clay and had a water holding capacity of approximately 23 percent, was determined. Vapam at the rate of 475 pounds per acre (125 gallons) was applied in water 6 and 12 inches deep in basins which were 10 feet square. Each treatment was replicated four times. Data on the control of the citrus nematode at different depths in the soil are presented in Table 4.

The citrus nematode was killed to the depth of approximately 3 feet when the Vapam was applied in 6 surface inches of water and to the depth of nearly 6 feet when Vapam was applied in 12 surface inches of water. Approximately 2 inches of water were required to wet 1 foot of the soil. Thus, the Vapam was carried downward in the water and apparently did not move farther than the water carried it.

Effect of Post-Irrigation Treatments on the Efficacy of Vapam: -- Frequently it is difficult to construct basin walls of sufficient height to retain large quantities of water and Vapam. A solution to this problem might be to apply the Vapam in a portion of the water required and then apply the remainder of the water later. The effectiveness of Vapam applied in this manner for control of the citrus nematode was determined in the following tests, which were made

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[3]Martin, J. P. 1950. Use of acid, rose bengal, and streptomycin in the plate method for estimating soil fungi. Soil Sci. 69:215-232.

[4]3.8 pounds n-methyl dithiocarbamate dihydrate per gallon, manufactured by the Stauffer Chemical Co.

Table 1. Mechanical analysis of sandy loam soil at the Citrus Experiment Station, Riverside[a].

Depth in inches	Clay (2 μ)	Silt (2-50 μ)	Sand (50 μ-2 mm)	Gravel (2 mm)
0 - 10	14.59	28.17	57.24	5.38
10 - 20	17.21	31.9	50.89	20.82
20 - 30	22.97	34.7	43.33	16.62
30 - 36	17.78	31.49	50.73	21.22
36 - 50	13.67	25.32	61.01	23.79
50 - 60	11.61	21.2	67.19	23.52

[a]pH 7.4 for 0-24 inch depth, 7.8 for 24-48 inch depth. Field capacity of soil approximately 11.0 percent.

Table 2. Mechanical analysis of fine sandy loam soil at Limoneira Company near Santa Paula[a].

Depth in inches	Clay (2 μ)	Silt (2-50 μ)	Sand (50 μ-2 mm)	Gravel (2 mm)
0 - 16	13.07	41.56	45.37	3.35
16 - 24	13.59	49.01	37.40	1.78
25 - 30	11.24	37.94	50.82	11.86
30 - 36	12.29	41.58	46.13	.80
36 - 42	19.43	62.66	17.91	.80
42 - 48	15.69	57.36	26.95	.18
48 - 53	11.51	52.30	36.19	.08
53 - 60	25.83	60.12	14.05	.74
60 - 64	15.04	38.07	46.89	2.12

[a]pH 7.1 for 0-24 inch depth, 7.8 for 24-48 inch depth. Average field capacity of the soil at 0-8 feet depth, 18.1 percent moisture.

Table 3. Control of the citrus nematode obtained with different concentrations and amounts of Vapam solutions.

Concentration of Vapam in water (ppm)	Depth of solution in basins (inches)	Pounds active ingredient per acre	Percentage control of the citrus nematode at depths indicated (inches)[a]			
			0-12	12-24	24-36	36-48
50	6	68.5	92	45	46	0
50	12	137	99	100	79	0
100	6	137	100	92	16	0
100	12	274	100	100	100	85
Untreated	6 inches water	0	0	0	0	0

[a]The figures are the average for two plots.

Notes: Moisture content when treated — 0-24 inch depth 7.0 percent; 24-48 inch depth 8.8 percent. Soil temperature at 12 inch depth 68° F. Field capacity approximately 11 percent moisture.

Table 4. Efficacy of 475 pounds of Vapam per acre for control of the citrus nematode in a loam soil.

Depth in feet	Average number of citrus nematodes per 50 ml of soil for the indicated treatments[a]		
	Untreated	Vapam applied in water	
		6 inches	12 inches
0 - 2	478	0	0
2 - 4	315	12	0
4 - 6	168	75	2
6 - 8	113	117	29

[a]Soil samples for determination of live citrus nematodes were taken 7 weeks after treating. The figures are the average for four plots.

Table 5. Effect of post-applications of water on control of the citrus nematode with Vapam.

Treatment	Average number of larvae per 50 ml of soil from different depths in feet[a]			
	0 - 1	1 - 2	2 - 3	3 - 4
Vapam 137 pounds per acre —				
In 4 acre-inches of water[b]	0	0	67	212
In 4 acre-inches of water plus 4 acre inches	0	0	0	0
In 4 acre-inches of water plus 8 acre inches	0	0	0	0
Untreated 8 acre inches of water	197	600	238	317

[a]Soil samples for determination of live citrus nematodes were taken 2 weeks after treatment. Average for four replications.
[b]137 pounds of Vapam in 4 acre inches of water is equivalent to 150 ppm

Table 6. Effect of adding water at different periods after applying Vapam on citrus nematode control.

274 pounds of Vapam per acre applied in depth of water indicated (inches)	Concentration of Vapam solution (ppm)	Number hours after treatment for addition of water 4 inches deep	Number of nematodes per 50 ml of soil at designated depths (inches)[a]			
			0-12	12-24	24-36	36-48
6	200	0	0	0	0	0
2	600	1	0	0	0	0
2	600	24	0	29	419	267
2	600	48	0	19	224	65
2	600	72	0	335	481	194
Control (6 inches of water)			637	1274	1222	550

[a]The figures are the average for two plots.
Note: Soil temperature at 12-inch depth 80° F.

Table 7. Control of the citrus nematode by Vapam sprinkled on the soil surface and then sprinkler-irrigated.

Treatment[a]	Average number of larvae per 50 ml of soil from indicated depths (inches)[b]			
	0-12	12-24	24-36	36-48
Vapam applied in 6 inches of water in basins	0	0	0	0
Vapam applied on surface and sprinkled after 5 to 10 minutes	0	13	162	360
Vapam applied on surface and sprinkled after 25 to 35 minutes	0	155	537	421
Untreated	1434	1838	1012	1188

[a]The Vapam was applied at the rate of 274 pounds per acre.
[b]The figures are the average for duplicate plots.

Table 8. Control of the citrus nematode by Vapam applied by different methods on a fine sandy loam soil.

Treatment[b]	Number of larvae per 50 ml of soil at indicated depths (feet)[a]			
	0 - 2[c]	2 - 4	4 - 6	6 - 8
Vapam 200 pounds per acre injected 7 inches deep plus 10 inches water by furrows	4	0	17	16
Vapam 300 pounds per acre injected 7 inches deep plus 10 inches water by furrows	6	6	14	5
Vapam 400 pounds per acre injected 7 inches deep plus 10 inches water by furrows	1	1	18	3
Vapam 300 pounds per acre injected 7 inches deep and cultipacked	26	66	29	6
Vapam 300 pounds per acre in 10 inches water by furrow	0	12	13	3
Vapam 300 pounds per acre in 10 inches water by sprinkler	1	7	21	1
Untreated	34	46	29	8

[a]The figures are the average for four replicated plots.
[b]Treated Dec. 14, 1955, sampled Feb. 13, 1956.
[c]LSD 1:99 = 33 for the 0-2 foot depth. Differences at the other depths are not highly significant.

on a sandy loam soil described in Table 1.

Vapam was applied at the rate of 137 pounds per acre in 4 surface inches of water in 12 metal basins, as illustrated in Figure 1. After 8 hours, when the Vapam solution had disappeared, water to a depth of 4 inches was placed in four basins and to a depth of 8 inches in another four basins. Eight inches of water also were added to four basins for a control. The numbers of citrus nematodes in the soil were determined after 14 days and are presented in Table 5.

The Vapam applied in 4 surface inches of water killed all of the citrus nematodes in the top 2 feet of soil and approximately 72 percent in the third foot. When either 4 or 8 inches of water were applied as soon as the Vapam solution had infiltrated into the soil, the citrus nematodes were killed to a depth of 4 feet or more. The additional water apparently carried the Vapam deeper into the soil to kill the nematodes in the third and fourth foot, but did not impair effectiveness near the surface.

Occasionally it might not be convenient to apply additional water as soon as the Vapam solution had entered the soil. Therefore, the effect of adding water at different periods of time after applying the Vapam solution was determined. The treatments were made in basins and in the same field as in the previous test. Vapam at the rate of 274 pounds per acre was applied in 13 gallons of water per basin, which was equivalent to water 2 inches deep. Then, water 4 inches deep was placed in the basins after periods of 0, 1, 24, 48, and 72 hours. Each treatment was made in duplicate. The number of citrus nematode larvae obtained from the soil 18 days after treating is presented in Table 6.

The citrus nematodes were killed to a depth of 4 feet when the Vapam was applied in 6 surface inches of water and also when the second increment of water was added 1 hour after applying the Vapam solution. However, when the water (4 inches deep) was applied 24, 48, or 72 hours after the Vapam solution, the nematodes in the first foot of soil were killed but in the second foot were only decreased in number. Thus, after 24 hours none of the chemical, or low concentrations, was carried downward with the water. This suggests that possibly the Vapam was absorbed on the soil, or had changed to insoluble compounds, or had escaped from the soil. The latter does not appear likely, since it was shown in the earlier tests that the Vapam did not move readily as a vapor in this soil. The soil temperature was 80° F at the 12-inch depth when the Vapam solution was applied. At lower soil temperatures increased control from adding water may occur over a longer period of time.

Nematocidal Properties of Treated Soil: -- Since only slight movement of Vapam in soil occurred 24 hours after applying, it appeared desirable to determine if the treated soil was toxic to nematodes after this time. Citrus nematode-infested soil was placed carefully in cylinders 12 inches in diameter. Then Vapam 1.4 gm in 3.8 liters of water was applied on the surface of the soil in each of four cylinders. This was sufficient solution to wet all of the soil. Equal amounts of the treated and of untreated infested soil were mixed and placed in quart jars 24, 48, 72, and 96 hours after treating. Two grams of orange roots infested with the nematode also were placed between layers of treated soil in jars after these periods. The jars were kept sealed 72 hours, then the roots and 50 ml aliquots of the soil were placed on funnels for determination of live citrus nematodes. Each treatment was replicated four times.

No citrus nematode larvae were obtained from the mixture of equal parts of treated and untreated soil, nor from the roots placed in contact with treated soil. From 50 ml of untreated soil and from 2 gm of untreated roots 1334 and 601 citrus nematode larvae were obtained, respectively. In this test soil treated with Vapam was highly toxic to citrus nematode larvae 24 to 96 hours after treating.

Vapam Applied Over the Surface of the Soil and Then Sprinkled: -- The efficacy of Vapam sprinkled over the surface of the soil and then water applied by sprinklers was tested for control of the citrus nematode. This type of treatment was tested on a sandy loam soil, similar to that described in Table 1. Vapam at the rate of 274 pounds per acre (139 gm in 2 gallons of water) was sprinkled evenly over the surface of tilled soil on plots 7 feet square. Sprinklers then were turned on 5 to 40 minutes after supplying the Vapam. The sprinklers were operated for 20 hours and delivered approximately 8 surface inches of water over the plots. However, after the first hour considerable run-off occurred. For comparison the same dosage of Vapam was applied in water 6 inches deep in the basins.

Vapam applied in 6 surface inches of water in basins killed the citrus nematode to a depth of 4 feet (Table 7). However, when Vapam was applied on the surface of the soil and then ir-

rigated by sprinkling, the citrus nematodes were killed in the first foot but decreased in number in the 12- to 48-inch depth. The poor control obtained below the first foot on the sprinkled plots apparently was due to rapid decomposition of the Vapam at the surface of the soil and the escape of volatile compounds to the atmosphere.

Vapam Applied by Injection and in Irrigation Water On a Fine Sandy Loam Soil: -- In certain of the above tests Vapam applied in water in basins controlled the citrus nematode satisfactorily. However, this method usually is not satisfactory when treating large areas or sloping land because of the difficulty of making level basins and of treating the ridges that form the basins. Thus, it appeared important to determine the efficacy of Vapam applied by other methods for control of the citrus nematode and parasitic soil fungi. Vapam was applied 1) by injection, 2) by injection followed by furrow irrigation, 3) by injection followed by sprinkler irrigation, and 4) in water by furrows and 5) in water by sprinklers.

On a fine sandy loam soil at the Limoneira Company near Santa Paula Vapam at doses of 200, 300, and 400 pounds per acre was injected and cultipacked. After approximately 24 hours shallow furrows were made and irrigated for 25 hours. It was estimated that approximately 10 acre-inches of water penetrated the soil. Vapam at 300 pounds per acre was injected and cultipacked. The same dose was metered into water at a uniform rate and applied by furrows over a 25-hour period and also in 10 surface inches of water by sprinklers over a 27-hour period (Fig. 3).

As shown in Table 8 the control of the citrus nematode obtained with these different Vapam treatments was unsatisfactory. Best control was with 300 pounds of Vapam per acre applied in water by furrows or by sprinkler irrigation, and by 400 pounds per acre injected 7 inches deep with shanks 12 inches apart followed by irrigation. In a previous test in this field (Table 4) Vapam applied at a rate of 475 pounds per acre in 12 acre-inches of water in basins killed the citrus nematode to a depth of 6 feet.

The number of fungi in the top 2 feet of soil was decreased approximately 56 percent by 300 pounds of Vapam per acre, applied in water by furrows or sprinklers. However, Phytophthora syringae (a cause of brown rot) was not killed by the treatments.

Vapam Injected and then Furrow-Irrigated at Riverside and Loma Linda: -- The efficacy of 300 and 400 pounds of Vapam per acre was tested on a sandy loam soil at the Citrus Experiment Station, Riverside. The Vapam was injected 6 inches deep, cultipacked, shallow furrows were made and then irrigated 2 to 4 hours after applying the chemical. Approximately 8 acre-inches of water was applied during a 24-hour period. Each plot was 24 by 100 feet and the treatments were replicated 4 times. The number of citrus nematodes at different depths in the soil was determined 2 1/2 months after treating.

The treatments consisting of 300 and 400 pounds of Vapam per acre killed 95 and 97 percent of the citrus nematode in the first foot of soil, respectively, and 74 percent in the second foot. The same treatments killed 65 and 70 percent of the non-parasitic fungi in the top 2 feet of soil, respectively, but neither treatment killed the brown rot fungus P. citrophthora. Control of both fungi and nematodes was unsatisfactory.

Vapam at the rate of 380 pounds per acre was injected into a deep sandy loam soil on the C. T. Norwood property near Loma Linda. Immediately, furrows were made and water was run for 51 hours on the four treated and nontreated plots.

Approximately 99 percent of the citrus nematodes and 66 percent of the non-parasitic fungi in the soil were killed to the depth of 4 feet in the soil.

Vapam Injected and Sprinkle-Irrigated at Corona and Fallbrook: -- Vapam at doses of 300 and 400 pounds per acre was injected 6 to 7 inches deep on a sandy loam soil on the J. Jameson Ranch near Corona. Between 2 to 6 hours after applying the Vapam the plots were sprinkle-irrigated for 24 hours. Approximately 8 surface inches of water was applied. Each plot was 12 by 80 feet in area and the treatments were replicated four times. The number of citrus nematodes in the soil at 1-foot increments to 4 feet deep was determined 5 months after treating.

The Vapam treatments killed between 31 and 91 percent of the citrus nematodes at the different depths.

The 300 pounds per acre treatment caused only slight reduction in the total number of fungi cultured from the soil, whereas the 400 pound dose decreased by 74 percent the number of fungus colonies. Two brown rot fungi, P. citrophthora and P. parasitica, were not controlled by these treatments.

On the Joe Strittmatter Ranch near Fallbrook Vapam was applied similarly except that the

plots were sprinkler-irrigated for 12 hours.

Between 82 percent and 100 percent of the lemon trees on the Vapam-treated and 100 percent on the non-treated plots were infected severely with P. parasitica 3 months after planting. The 300 and 400 pounds treatments killed the citrus nematode to depths of 2 and 4 feet in the soil, respectively.

Vapam Applied by Sprinklers at Upland and Vista: -- On a gravelly soil on the F. Gingerich Ranch near Upland, Vapam was applied at doses of 200 and 400 pounds per acre by sprinklers. The Vapam was applied in 5 surface inches of water during a 12-hour period. The water entered the soil readily and there was no run-off. Soil samples for determination of fungi and the citrus nematode were taken 2 and 5 months after treating, respectively.

The soil in this field contained approximately 2 percent clay, 12 percent silt, and 86 percent sand and gravel with many large rocks tossed in. It is of interest that 5 surface inches of water containing 177 ppm of Vapam (200 pounds of Vapam per acre in 5 acre-inches of water) applied by sprinklers controlled the citrus nematode in the first foot of soil only. However, when 5 surface inches of water containing 354 ppm of Vapam were applied (400 pounds of Vapam in 5 acre-inches of water) the nematode was controlled to a depth of 3 or more feet, but not the brown rot fungus. The number of fungus colonies isolated from the top 2 feet of soil was decreased by 48 and 95 percent with the 200 and 400 pound doses, respectively.

On the E. T. Sorenson Ranch near Vista, Vapam was applied on two large plots at the rate of 400 pounds per acre in 5 acre-inches of water by sprinklers during 15 hours.

Four months after planting 13 percent and 64 percent of the lemon trees on the treated soil and 100 percent on the non-treated soil were infected severely with P. parasitica. All of the citrus nematodes were killed and the number of colonies of non-parasitic fungi were decreased 89 percent in the 0- to 4-foot depth of soil.

Vapam Applied in Basins at Corona and Rubidoux: -- Vapam at the rate of 400 pounds per acre was applied in basins in 5 to 6 surface inches of water on the Campbell Sloop Ranch near Corona and on the Tully Ranch near Rubidoux. In both groves the brown-rot fungus P. parasitica was controlled by the Vapam treatments. In the grove near Corona, 99 of 100 trees planted on treated soil grew satisfactorily the first year, and only 20 out of 2000 planted on non-treated soil survived. At Rubidoux, 40 lemon trees on the Vapam-treated soil remained free of Phytophthora while 10 of 11 trees on non-treated soil became infected with either P. citrophthora or P. parasitica. The non-treated soil in these groves was free from or infested slightly with the citrus nematode.

Effect of pH of the Soils: -- All of the soils on which the tests were conducted were slightly alkaline, except two. At the 0- to 24-inch depth a range of pH 7.1 to pH 7.4 occurred for the alkaline soils. The soils near Fallbrook and Loma Linda were pH 5.2 and pH 6.7 near the surface, respectively. At the 24-inch to 48-inch depth the pH of all of the soils was 0.4 to 1.1 higher than near the surface. In these tests soil pH had no apparent effect on the control of the citrus nematode with Vapam.

Period to Wait Before Planting after Treating: -- Sandy loam soils from two fields that had been treated with 400 pounds of Vapam per acre were toxic to small sweet orange seedlings planted 2 weeks after treating, but not after 3 weeks. It appears that citrus trees may be planted safely 3 weeks after treating with Vapam on most soils.

DISCUSSION AND SUMMARY

The citrus nematode was effectively controlled when 274 to 475 pounds of Vapam per acre were applied in 6 to 12 surface inches of water in basins. The low doses were effective on sandy loams and the high doses on loam soils. The treatments also decreased the number of saprophytic fungi in the soil. Both Phytophthora citrophthora and P. parasitica were killed by 400 pounds of Vapam per acre applied in 5 to 6 surface inches of water in basins on sandy loam soils. Control of the citrus nematode and the brown-rot fungi was unsatisfactory when Vapam was injected into the soil and cultipacked, injected into the soil and irrigated, applied on the surface and sprinkled, or applied in water by furrows. On a sandy loam soil and a gravelly soil 400 pounds of Vapam per acre applied in 5 surface inches of water by sprinklers controlled the citrus nematode, but not P. parasitica or P. syringae. However, on a fine sandy loam soil control of both fungi and the citrus nematode was unsatisfactory when Vapam was

applied by sprinklers.

Vapam moved through a sandy loam and a fine sandy loam soil chiefly in water, and was sorbed on the soil or changed to insoluble compounds within 24 hours. It then did not move appreciably through the soil; however, it remained nematotoxic. Lear[5] studied the nematocidal properties of Vapam for control of the root-knot and sugar-beet nematodes. He reported that a 4 ml dose diffused 6 to 8 inches laterally and only 4 inches vertically.

The acidity of soils within the range pH 5.2 to pH 7.8 apparently did not affect the control of the citrus nematode and Phytophthora spp. with Vapam.

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[5]Lear, Bert. Results of laboratory experiments with Vapam for control of nematodes. Plant Dis. Reptr. 40:847-852. 1956.

SOIL TREATMENTS FOR CONTROL OF THE RED STELE DISEASE OF STRAWBERRIES[1]

Walter F. Jeffers[2]

Summary

Under controlled conditions chloropicrin was very effective in eradicating Phytophthora fragariae from infested soil. Thiram (Arasan) and CBP also were effective but caused severe plant injury at the concentrations used. Several fungicidal materials including nabam (Dithane D-14) and captan (Orthocide) were ineffective. Under field conditions none of the materials resulted in complete eradication of the red stele fungus, but methyl bromide and chloropicrin, at relatively low rates, gave a high degree of control.

Phytophthora fragariae Hickman, the causal agent of the red stele disease of strawberry, is widely distributed in moist soils in much of the strawberry-growing area of the northern United States. One feature that accounts for severity of the red stele disease is the ability of the pathogen to remain viable for many years in soil even in the absence of host plants. Apparently this ability to withstand unfavorable conditions is due to the resistant nature of oospores of the organism.

Aside from the work of Stoddard (1, 2) there is little published information on the effectiveness of chemicals for eradication of the red stele fungus from infested soils. Some appreciation of this problem was experienced by the author in 1941 when an attempt was made to eliminate P. fragariae from a small, low area in a large field. This spot, of about 3,700 square feet, was drenched, in June, with 8 gallons of water containing 1 quart of Elgetol. The following fall 25 strawberry plants (Blakemore variety) were planted near the center of this area and when examined in the spring 15 of them showed typical symptoms of red stele.

Later, a more extensive test of chemicals for eradication of P. fragariae was conducted under controlled conditions. Glazed nutrition crocks (2-gallon size) were filled to about 2 inches from the top with soil known to be heavily infested with race 1 of this organism, and randomized in 3 replications on a raised greenhouse bench. On December 12, 1952 chemicals were applied to the crocks of soil and 7 days later five susceptible strawberry plants (a selection of Suwanee x Midland) were planted in each crock. In order to allow the plants to make new root growth the temperature was maintained at about 70° to 75° F for 30 days. Then the greenhouse was allowed to cool to a range of approximately 35° to 55° to provide a favorable temperature for zoospore production and subsequent infection. To further facilitate infection soil moisture was maintained at a high level. On January 2, 1953 some plants were dead as a result of injury from the chemicals and these were replaced with more plants of the same clone. On April 1, 1953 the plants were removed and examined for red stele and for evidence of phytotoxicity. A summary of this experiment is presented in Table 1.

Although the concentration of several materials was sufficient to cause considerable damage to roots of the strawberry plants the pathogen still was able to cause much infection. Even copper sulfate and formaldehyde at the rates at which they were applied did not prevent infection under conditions of this experiment. Thus it is evident that P. fragariae is quite resistant to some fungicidal materials. It is of interest that some gaseous materials were effective in eradicating this pathogen from soil.

Another experiment was conduced concurrently with this one. Its purpose was to determine whether chemical treatment during the period when infection was occuring would be more effective than treatments made while the pathogen was dormant. Susceptible strawberry plants were planted, on December 15, 1952, in infested soil in 2-gallon crocks that were randomized in 4 replications on a greenhouse bench. The temperature was maintained at about 70° to 80° F, until January 18, 1953, to produce new root growth and to prevent any infection by the red stele pathogen. On January 18, 1953 the temperature was lowered to a range of

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Table 1. Results from soil treatments for eradication of Phytophthora fragariae.

Materials and amounts applied on Dec. 12, 1952*		Phytotoxicity		Number of plants	
		Jan. 2, 1953	Apr. 1, 1953 (after replanting Jan. 2, 1953)	Alive Apr. 1 1953 of 15 planted	With red stele
Chloropicrin	3.0 ml	All plants dead	None observed	15	0
D-D	5.0 ml	Severe	Severe	3	0
Ethylene dibromide 100%	2.0 ml	Severe	All plants dead	0	—
Formaldehyde 37.5%	29.0 ml	All plants dead	Slight	15	15
CBP 55 (1-dichloro-3-bromopropane 55%)	5.0 ml	Severe	Severe	11	0
Dithane D-14 (nabam)	250.0 ml	All plants dead	Severe	8	4
Dithane Z-78 (zineb)	72.5 g	All plants dead	Severe	12	1
Orthocide 406 (captan)	72.5 g	Severe	Severe	12	12
Vancide 51	10.0 ml	None noted	Severe	15	13
Vancide 51	100.0 ml	All plants dead	Severe	4	0
Arasan (thiram)	72.5 g	None noted	Severe	15	0
Mercuric chloride	1.0 g	Severe	Severe	3	2
Copper sulfate	25.2 g	All plants dead	Severe	11	3
Steam	15 pounds/2.5 hours	None noted	None noted	15	0
No treatment				15	14

*Figures refer to amount of material applied to approximately 32 pounds of soil in a glazed 2-gallon crock. Crocks treated with fumigants were covered with plastic for 48 hours at about 70° F. Liquids were made up to 1 liter with water and dry materials were added to 1 liter of water. Crocks receiving liquid treatments, except Dithane D-14 and Dithane Z-78, were plugged for 48 hours. There were 3 replications of each treatment, i. e., 3 crocks each containing 5 plants.

Table 2. Effectiveness of chemicals applied to soil during early stage of infection in controlling red stele disease of strawberry.

Materials tested	Rate of application to each crock of soil*	Phytotoxicity	Number of plants: Alive of 20 planted	Number of plants: Showing red stele symptoms
Orthocide 406 (captan)	1 g per liter of water	Severe injury	20	20
Vancide 15	15 ml in 985 ml water	Severe injury	10	8
Parzate Liquid (nabam)	15 ml in 985 ml water	Severe injury	20	2
Dithane D-14 (nabam)	15 ml in 985 ml water	Severe injury	15	1
Tri-basic Copper Sulfate ($CuSO_4$ $3Cu(OH)_2$) 53% Cu	1 g per liter of water	Severe injury	20	16
Water only	1 liter	Slight water damage	20	20

*Crocks were of 2-gallon size and each contained about 32 pounds of soil.

Table 3. Application of soil fumigants for control of red stele of strawberry.

Treatment*	Amount per 100 square feet	Number of plants showing red stele of 100 planted
D-D	400 ml	33
Chloropicrin	271 ml	4
Dowfume W-40 (ethylene dibromide 40%)	400 ml	66
CBP 55	400 ml	42
Methyl bromide	1 pound	1
No treatment		72

* Methyl bromide was applied by use of a pressurized container which released the gas under a plastic cover allowed to remain in place for 48 hours. All other materials were applied with a hand injector to a depth of about 6 inches. The chloropicrin plots were covered with plastic for 48 hours following application. No cover was used on the other plots.

about 40° to 60° to provide a favorable condition for production of zoospores and subsequent infection. On January 31, 1953 the first application of chemicals was made, and a second treatment was made on January 21, 1953. The drainage hole in the bottom of each crock was plugged and the liter of material that was applied each time appeared to be sufficient to saturate the soil thoroughly. It was originally intended to apply the chemicals at 10-day intervals for 3 months. However, so much plant injury was evident after the second application that no further treatments were made. On April 3, 1953 the plants were removed from the soil and examined for red stele. A summary of this experiment is presented in Table 2.

It was evident that there was much chemical injury to the plants as well as considerable damage due to lack of soil aeration. However, despite the amount of fungicide applied some infection was found in plants from each treatment. The nabam materials showed a greater degree of control than the other materials.

To test soil fumigants for eradication of the red stele fungus under field conditions various treatments were applied on September 10, 1953. An area in a field was selected in which race 2 of P. fragariae had caused severe injury to the Temple variety of strawberry the previous spring. There were 4 replications of each of 6 treatments. Each block was 10 by 10 feet and was separated from adjoining blocks by a ditch about 6 inches deep and 12 inches wide. On November 13, 1953, 25 plants of a susceptible strawberry selection were planted in the central area of each block. On April 7, 1954 all plants were removed and examined for red stele infection. Treatments and amount of infection are shown in Table 3. Apparently methyl bromide and chloropicrin could be used successfully to eradicate the red stele organism. However, dosages somewhat greater than those used in this experiment appear to be necessary.

Literature Cited

1. STODDARD, E. M. 1951. A chemotherapeutic control of strawberry red stele. Phytopathology 41: 34.
2. STODDARD, E. M. 1951. Control of strawberry red stele by chemotherapy. Phytopathology 41: 858.

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IN-THE-FURROW APPLICATION OF FUNGICIDES FOR THE CONTROL OF COTTON SEEDLING DISEASE -- DAMPING-OFF AND NUB-ROOT[1]

Marvin D. Whitehead and Norman E. Brown

Summary

Ten fungicides were tested separately and in certain combinations when applied in the furrow as possible protectants against the cotton seedling disease complex. Four replicated plots, non-inoculated and inoculated with a group of fungal cultures known to be seedling disease incitants, were tested over a two-year period. The fungicide treatments captan, nabam, PCNB plus nabam, and nabam plus zinc sulfate gave higher plant populations that produced yields more than two-fold that of the untreated plots. Significant differences in yield at first picking followed by significantly larger total yield increases were obtained. Highly significant differences were noted in the reduction of nub-root symptoms.

It has been generally recoginzed that cotton seedling disease of the damping-off phase is incited by a complex of soil-borne microorganisms. In 1955 Stewart and Whitehead (4) described the mature plant phase of seedling disease, nub-root, incited by the same microorganism complex, as a disease having a greater yield-reducing potential than the damping-off phase. In comparison, the yield loss from nub-root to the Missouri cotton crop was estimated as 7.5 percent in 1955 and 5.0 percent in 1956, with damping-off taking an additional 2.5 percent.

Fungicidal seed treatment before planting has failed to give sufficient protection from this disease. The in-the-furrow method of application of fungicides has stemmed from a laboratory testing technique described by Arndt in 1953 (1). Cooper in 1954 (2) reported highly significant cotton stand increases (almost two-fold) from in-the-furrow applications of captan and thiram, while a group of other fungicides gave significant increases. He stated that the method showed promise in the development of control for cotton stand failures incited by soil-inhabiting organisms. The application of fungicides to the furrow for the control of the cotton seedling disease complex was first tested in a United States Southern regional testing program during the 1954 season. Results reported at the annual meetings of the Cotton Disease Council have been erratic and variable because of chance presence of environmental conditions favorable for seedling disease development (3).

METHODS AND MATERIALS

The tests herein reported were conducted at Sikeston, Missouri on sandy loam soil in an area of Mississippi River alluvial soil located at a point near the northern limit of cotton production in the United States. The in-the-furrow fungicides were tested in treatments uninoculated and inoculated with cultures of fungi known to incite seedling disease. The inoculum was cultured on a grain sorghum medium (5). Prior to planting, the seed of Delfos 9169 variety was inoculated by mixing the grain sorghum cultures with seed. To test the fungicides against some of the different genera of fungi of the seedling disease complex in the 1955 tests, the seed of block 1 in 2-row plots were inoculated with a mixture of 10 cultures of Rhizoctonia solani, block 2 with a mixture of 10 cultures of Fusarium moniliforme, F. chlamydosporum, and Fusarium spp., and block 3 with a mixture of 10 cultures of Pythium debaryanum and P. ultimum. In the 1956 planting, four replications of 4-row plots were planted. The fungicide treatments were applied to uninoculated seed and to seed that had been inoculated with a mixture of cultures of Rhizoctonia solani, Fusarium moniliforme, F. chlamydosporum, Fusarium spp., Pythium debaryanum, and P. ultimum.

Mechanically delinted Ceresan-treated (2 ounces per 100 pounds) seed were used in all

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Table 1. Percentage emergence and survival of cotton plants 44 days after planting and corresponding yield in pounds of lint cotton per acre of 1955 in-the-furrow fungicide tests at Sikeston, Missouri.

Fungicide applied per acre	Rhizoctonia inoculated		Fusarium inoculated		Pythium inoculated		No inoculation		Average
	Stand*	Yield	Stand	Yield	Stand	Yield	Stand	Yield	Yield
Check	10.3	454	16.3	580	41.9	661	43.3	753	612
Ceresan S, 1 pt.	9.3	485	9.5	454	27.3	575	20.3	634	537
Ceresan M2X, 1 lb.	9.1	481	9.1	341	16.7	621	14.8	475	479
Panogen 15, 5 lb.	5.9	423	7.8	343	15.0	594	7.8	345	426
PCNB (Terraclor), 5 lb.	14.6	412	17.3	643	21.6	659	44.8	647	590
Captan, 75% WP, 5 lb.	18.0	567	14.6	605	39.9	626	56.6	628	606
Merbam, 0.25 lb.	11.4	490	19.9	611	34.0	670	44.0	594	591
Gallotox, 5 pt.	15.0	559	17.1	525	27.9	561	33.8	596	560
Mathieson 363, 5 lb.	9.9	584	14.2	630	44.4	697	41.6	724	659
PCNB, 5 lb. plus Merbam, 0.25 lb.	8.5	467	18.4	632	37.4	666	45.9	695	615
PCNB, 5 lb. plus Captan, 2.5 lb.	11.0	485	15.0	571	33.0	636	53.1	643	584
Nabam, 5 qt.	51.6	760	57.3	774	41.6	739	57.3	666	735
Nabam, 5 qt. plus $ZnSO_4$, 0.5 lb.	52.8	722	58.3	780	52.2	722	60.2	644	717
Nabam, 5 lb. plus PCNB, 5 lb.	48.8	640	50.0	718	49.5	799	61.5	757	728

*Count from 100 ft. of row from seeding rate of 4.73 seed per foot of row.

Table 2. Emergence and survival in average number of plants per foot of row 46 days after planting and the corresponding yield in pounds of seed cotton per acre of 1956 in-the-furrow fungicide tests at Sikeston, Missouri.

Fungicide applied per acre	Plant Stand*		Yield			
			inoculated		non-inoculated	
	inoculated	non-inoculated	First picking	Total	First picking	Total
No treatment	0.45	1.50	238	1076	317	1505
Captan 50% W.P., 5 pounds	3.93	6.12	977	2429	1089	2191
Panogen 15, 2.5 pints.	2.52	3.87	601	2006	825	2284
O-nitrochlorobenzene, 2 pounds	0.42	1.02	20	224	297	706
PCNB (Terraclor), 4 quarts	0.45	1.55	145	1060	370	1630
Nabam, 5 quarts	4.66	6.18	1030	2330	1030	2185
Nabam, 4 quarts plus PCNB, 4 quarts	3.79	6.84	977	2303	1023	2185
Nabam, 4 quarts plus $ZnSO_4$, 0.5 pounds	3.76	6.35	1109	2442	997	2219
L.S.D. .05	0.36	0.61	185	317	271	284

*Stand count from 200 foot of row from seeding rate of 10 seed per foot of row.

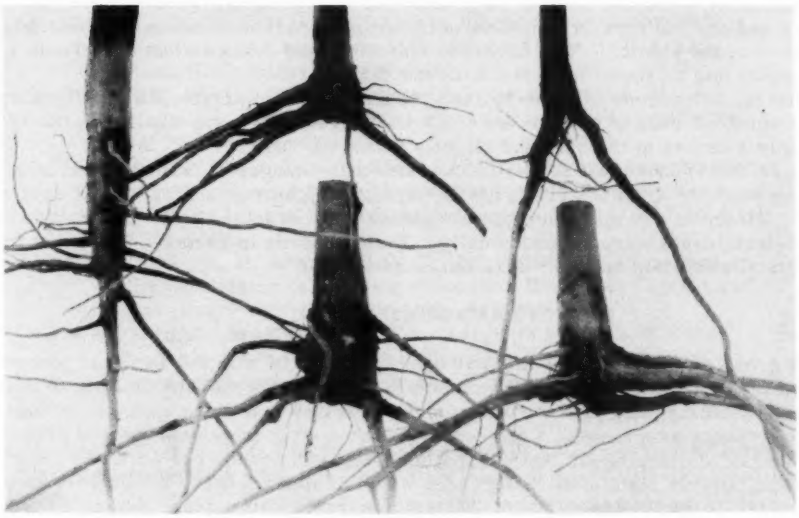

FIGURE 1. Nub-root of cotton--one healthy, four diseased. Adventitious root development in the upper soil layer is made possible by sufficient available moisture.

Table 3. Percentage of plants showing nub-root symptoms in fungicide tests at Sikeston, Missouri, 1956.

Fungicide applied per acre	Inoculated	Non-inoculated
No treatment	37.0	24.0
Captan 50% W. P., 5 pounds	6.0	5.5
Panogen 15, 2.5 pints.	18.7	14.0
O-nitroclorobenzene, 2 pounds	47.5	40.0
PCNB (Terraclor), 4 quarts	28.7	22.7
Nabam, 5 quarts	12.0	8.0
Nabam, 4 quarts plus $ZnSO_4$, 0.5 pounds	6.5	4.5
Nabam, 4 quarts plus PCNB 4 quarts	5.5	5.5
L.S.D. .05	10.7	8.0
.01	14.6	10.8

treatments. The fungicide materials were applied in the furrow at planting time by spraying at a rate of 10 gallons of water per acre at 20 pounds per square inch pressure from two nozzles (one cone and one fan type) mounted directly over the furrow between the seed tube and covering device of the planter. The fungicide materials and rates as listed in Table 1 were used in the 1955 tests and as listed in Table 2, in the 1956 tests.

In the 1955 tests, 300 pounds of 0-20-20 and 100 pounds of ammonium nitrate fertilizers were applied per acre. A band of 200 pounds of 13-13-13 fertilizers was applied in the 1956 tests approximately 2 inches to the side and slightly below the seed level.

Stand counts as emergence and survival were made after danger of further stand loss was considered to have passed. Counts were made 44 days after planting in 1955 and 46 days after planting in 1956. Minimum and maximum air temperature records and soil temperature records at 1- and 3-inch levels were recorded daily. Yield records in pounds per acre at the first picking and total plot yield on a per acre basis were made.

RESULTS AND DISCUSSION

The selected group of seedling-infecting pathogens was mixed with the seed and placed in the seed furrow at planting time to insure the close proximity of the disease incitant to the seed and developing seedling, and to place each fungicide tested in a more uniform pathogen-host sphere. Stand counts as emergence and survival (taken after it was considered probable that most of the danger of damping-off had passed) and yields in pounds per acre are listed in Table 1 and Table 2. Highly significant differences were obtained between treatments as to stand count and yields. In the 1956 experiment the fungicide treatments captan, nabam, PCNB plus nabam, and nabam plus zinc sulfate gave increased stands that carried through to yields more than two-fold that of the untreated plots. The differences obtained in the 1955 test for nabam, nabam plus zinc sulfate, and nabam plus PCNB were of the same magnitude and closely correlate with those obtained in the 1956 season.

Each of the different blocks of the 1955 experiment were inoculated with a group of cultures of different genera of fungi (Table 1) to give some indication of specificity of some of the fungicides to a certain group of fungi of the seedling disease complex. The uninoculated block was used to check the control of the naturally occurring disease complex.

The organic mercury treatments with Ceresan and Panogen in the 1955 experiment reduced germination. The seed used had been pretreated with Ceresan and the additional application of the mercury compounds to the seed furrow apparently resulted in toxicity. Panogen, when reduced to one-half the 1955 dosage in the 1956 experiment, resulted in significant stand and yield increases over the check.

The significant differences in yield at first picking are of economic consideration. Apparently the plants were given an advantage at the period of early growth. This early healthy start apparently promoted early fruiting and allowed the plant to produce a larger percentage of the crop at the first picking when quality and prices are higher. Another factor in maturity is the fact that cotton fruits earlier when the populations are higher.

All plants in the 1956 tests were pulled after harvest to check root development. The percentage of plants showing nub-root symptoms (Fig. 1) (no tap root or fragmentary root development) is presented in Table 3. Highly significant differences in root development were noted in both the inoculated and uninoculated tests.

The relatively short growing season for cotton in Missouri requires that it be planted at the earliest possible date, which means planting in cold, wet soil (Table 4) that may remain cold for a considerable period. These circumstances give to Missouri's location at the northern limit of cotton production, with all the cotton acreage of the State concentrated in seven counties of the Mississippi River Delta, edaphic and climatic characteristics especially favorable to expression of seedling disease and especially suitable for cotton seed treatment studies.

Literature Cited

1. ARNDT, C. H. 1953. Evaluation of fungicides as protectants of cotton seedlings from infection by Rhizoctonia solani. Plant Dis. Reptr. 37: 397-400.
2. COOPER, W. E. 1954. The seed furrow application of fungicides to control cotton stand failures. Phytopathology 44: 331. (abstract)
3. RANNEY, C. C. and L. S. BIRD. 1956. Greenhouse evaluation of in-the-furrow fungicides at two temperatures as a control measure for cotton seedling necrosis. Plant Dis. Reptr. 40: 1032-1039.
4. STEWART, R. B. and MARVIN D. WHITEHEAD. (Nub-root -- the expression of seedling disease in the mature cotton and flax plant.) Phytopathology 45: 413-416.
5. WHITEHEAD, MARVIN D. 1955. Sorghum grain, a medium suitable for the build-up of inoculum for studies of soil-borne and certain other fungi. Phytopathology (In Press)

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THE PEANUT STEM ROT PROBLEM AND A PRELIMINARY REPORT ON INTERRELATIONS OF "NON-DIRTING" WEED CONTROL AND OTHER PRACTICES TO STEM ROT AND YIELD OF PEANUTS[1]

Kenneth H. Garren and George B. Duke[1]

Abstract

The stem rot of peanuts caused by Sclerotium rolfsii has been under investigation as a major problem of peanut production in the United States and elsewhere for 40 years. A summary of the work on this disease is presented as an introduction to a report on results obtained to date from a long-range study of stem rot now underway at Holland, Virginia in cooperation with the Virginia Agricultural Experiment Station. In 1955 an experiment with Virginia Bunch variety 46-2 peanuts compared "non-dirting" weed control (weed control by cultivation such that no soil was thrown onto the peanut plants) with "dirting" weed control (weed control by cultivation such that soil was thrown onto the peanut plants to smother weeds and grass). The plots for the non-dirting weed control received a pre-emergence treatment of the herbicide DNBP. This experiment also compared deep covering of winter cover crop and other organic matter making up the surface litter with shallow and incomplete covering of surface litter. The experiment was repeated in 1956. In both years infection was less and yield greater with the non-dirting weed control than with the dirting weed control. The differences were statistically significant at the 1% level. The 1956 results were in favor of deep covering as compared with shallow covering. In 1956 additional results from a test with Dixie Spanish variety peanuts suggested that the herbicide DNBP may have some beneficial effects independent of whether soil is thrown onto the peanut plants, and that a drench of PCNB also tends to be beneficial in reducing both above-ground infection and damage to pods and kernels.

INTRODUCTION

In 1917 McClintock (13) described a "wilt" of peanuts (Arachis hypogaea L.) caused by a soil-borne fungus, Sclerotium rolfsii Sacc. In 1920 Wolf (19) described a pod rot of peanuts caused by the same fungus. S. rolfsii attacks many cultivated and wild plants in warmer regions. Since it causes a soil-line rot of both stems and "pegs" (stalks of the pods) as well as a rot of the pods of peanuts it is a major pest of this crop plant. Higgins (12), in 1927, found that cells of plants attacked by S. rolfsii were killed before the hyphae penetrated them and that in the dead cells and the clinging mat of hyphae the oxalic acid content was high. Thus the damage caused by S. rolfsii is not centered in the vascular tissues, and "stem rot" seems a more specifically descriptive name than "wilt" for the disease which it causes.

Until recently all attempts at controlling stem rot of peanuts were discouraging. As is usual for a disease of this type the two main lines of investigation were a search for resistant varieties and a study of rotation practices. McClintock (13, 14) thought that Virginia runner varieties of peanuts were practically immune from or highly resistant to S. rolfsii, and in the Philippines Reyes (15) reported resistance in progeny of some crosses. These reports of resistance were not substantiated by subsequent reports and by 1953 (16) other reports of resistance which also apparently could not be substantiated by subsequent tests had appeared. Perhaps some varieties of the runner type of peanuts are less susceptible or more tolerant than are Spanish and most other bunch varieties of peanuts, but the confirming data have not appeared in the literature. Of the crops commonly used as alternates in peanut rotations only corn and cotton are resistant to S. rolfsii. Nevertheless stem rot is often a serious problem in Virginia where most peanuts are grown in 2-year rotations with corn. A survey by Garren (9) indicated that stem rot incidence on peanuts is sometimes high following cotton, and the

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surveyor attributed this unexpectedly high incidence of stem rot on peanuts immediately following cotton to the ability of S. rolfsii to grow vigorously on the debris usually left in cotton fields.

In 1921 Tisdale (18) recommended early plowing of rice fields infested with S. rolfsii to promote both decay of organic matter and germination and exhaustion of sclerotia. There were frequent expressions of a theory that effective control of leaf spot of peanuts caused by Cercospora spp. can prevent late-season accumulation of sufficient organic matter to support rapid growth of S. rolfsii. A 2-year study by Garren (9, 10) compared leafspot and stem rot development in peanuts dusted with ferbam, ziram, copper-sulfur, and sulfur and found no apparent effect on the amount of either disease. The ability of S. rolfsii to grow on plant debris and survive without contacting a living host received little attention until Boyle (1, 2) began his investigations. He found (2) that adding soybean debris to soil naturally infested with Rhizoctonia spp. and artificially inoculated with S. rolfsii resulted in an increase in both incidence and severity of infection on hypocotyls and roots of peanuts grown in the soil. Infection was also increased when trash of either cotton or corn was added, but to a less degree.

Another aspect neglected until recently was first noted in 1919 by Taubenhaus (17) who discussed the role of sclerotia of S. rolfsii in infection and suggested as a control measure deep plowing to bury sclerotia. On the other hand, it is common knowledge that covering the base of peanut plants with soil increases the amount of stem rot, but the degree of increase has not been determined. Farmers in general throw some soil around the base of peanuts as a means of controlling weeds and grass, and many farmers use this as their principal or only method of weed control. Some resort to "heavy dirting" when weather conditions delay cultivation and hoeing. Almost all peanut growers "bed" their crop late in the season by piling soil around the base of the plants.

Some work in keeping with newer trends has been done on peanut stem rot. Cooper (7) used drenches of various soil fungicides and found PCNB (pentachloronitrobenzene), captan, and thiram promising. Chappell and Miller (5) noted more vigor for peanuts in herbicide-treated areas and found herbicides effective in the laboratory against several fungi including S. rolfsii. They concluded that the use of herbicides may influence disease development but not that the ones tested actually acted as fungicides in the field. At least one herbicide, one of the dinitro-amine group, proved effective as a soil fungicide on a crop plant other than peanuts (4).

The bunch peanut variety "N. C. 2" is a result of modern plant breeding techniques. Cooper (6) reported that N. C. 2 has a greater tolerance to stem rot than the general farmers' stock of Virginia Bunch peanuts.

In 1952 Boyle (1) put together a theory as to how stem rot may be controlled and reported encouraging results from a preliminary test of it. This theory calls for (a) plowing to provide deep burial of organic litter on the surface of the soil; (b) control of weeds without throwing soil around the base of the plants; and (c) effective control of leaf spot. He reported, in personal communications, that from 1953 to 1955 drought prevented effective testing of the theory, but in 1956 Boyle and Hammons (3) reported further encouraging results.

Cooper (8) reported in 1956 preliminary results of a long-range rotation study. There was 19 percent infection by S. rolfsii in continuous peanuts, as compared with 8 percent in peanuts alternating with soybeans, and 4 percent in peanuts alternating with wheat interplanted with such crop plants as cotton and corn. There was an average infection of only 2 percent in peanuts in 3- and 4-year rotations.

Thus some helpful information was accumulated during the years when attempts at controlling stem rot gave discouraging results. In 1956 Cooper (8) summarized this progress in a report which recommended crop rotation, flat cultivation instead of late-season bedding, leaf spot control, and the use of more tolerant varieties.

In the spring of 1955 an experiment was started at Holland, Virginia to determine effects on development of stem rot of peanuts of both deep-covering of organic litter and sclerotia and of weed control without throwing soil onto the base of the plants. The study was expanded in 1956 to include inquiry on the fungicidal action of the herbicide DNBP (4, 6, dinitro ortho secondary butylphenol) and the effectiveness of the fungicide PCNB. This report is on the work in Virginia to date and it considerably enlarges on a brief report (11) previously presented on this same study.

PROCEDURE

The cropping history of the area used for the work in 1955 was: corn 1952, peanuts 1953, corn 1954, ryegrass winter of 1954-1955, and the experimental peanuts in 1955. The cropping history of the area used for the work in 1956 was: peanuts 1952, soybeans 1953, milo 1954, rye and lespedeza 1955, ryegrass winter of 1955-1956, and the experimental peanuts 1956.

A split-plot experiment, designated "A", was set up first in 1955 with two plots (I, deep covering of surface litter and sclerotia; II, shallow covering of surface litter and sclerotia) each with two subplots (1, "Non-dirting" weed control; 2, "dirting' weed control). The "non-dirting" weed control was by cultivation such that no soil was thrown onto the rows of plants and DNBP herbicide was always used in connection with this treatment in experiment "A". No herbicide was used in connection with the "dirting" method of weed control in experiment "A", and sufficient soil was thrown onto the rows of peanut plants to smother weeds and grass.

In 1956 this experiment was planted with farmers' stock seed of Virginia Bunch variety 46-2 peanuts and it consisted of 4 replications of 8 plots of 0.044 acre each. The deep-covering plots were plowed with two 12-inch bottom moldboard plows with coulters and jointers. The shallow covering plots were disked 6 times with a disk harrow and were then worked thoroughly to a depth of 8 inches with a spring-tooth harrow. The drill area of the non-dirting subplots was rolled in bands 16 inches wide at planting and 3 days after planting these subplots were treated with DNBP at the rate of 3 gallons of 53% active substance per acre.

The middles of the non-dirting subplots were cultivated 3 times. Cultivation ended in early August. Dirting of plants was prevented by attaching shields to the cultivator for the first cultivation and by using smaller sweeps for the second and third cultivations. These subplots were not hoed and the larger weeds were removed by hand. Early in the season the dirting subplots were rotary-hoed twice. The middles of these plots were cultivated at the time the non-dirting subplots were cultivated, but shields were not used and the sweeps were set so as to throw sufficient soil onto the base of peanut plants to control weeds and grass in the row.

All subplots were machine-dusted, the first time with sulfur plus 5% DDT and thereafter twice with sulfur.

Experiment "A" was repeated on another site in 1956. The 1956 experiment varied from the 1955 experiment only in the following minor respects: In 1956 the seed were of the same variety as in 1955 but they were from a more highly selected stock. In 1956 there were 8 replications of 4 plots of 0.017 acre each. In 1956 one 18-inch bottom plow was used instead of the two 12-inch bottom plows used in 1955.

A second experiment, designated "B", set up in 1956, differed from experiment "A" in that the non-dirting and dirting methods of weed control were both used with and without a herbicide. A treatment involving PCNB was also added. Experiment B had 2 plots (I, peanut hay trash added to surface of plots; II, no trash added to surface of plots) each with 5 subplots (1, non-dirting weed control with herbicide; 2, non-dirting weed control without herbicide; 3, dirting weed control with herbicide; 4, dirting weed control without herbicide; 5, dirting weed control without herbicide but with PCNB).

Experiment "B" had 4 replications with 10 0.008-acre plots per replication. Selected Dixie Spanish variety peanut seed was used. In early March loose peanut hay was spread to a depth of 2 inches on 4 of 8 blocks of this experiment, and thereafter all of the area received the same pre-planting treatment of disking and spring-tooth harrowing that the shallow covering plots of experiment "A" received. Thus while experiment "B" largely replicated experiment "A" it also included an inquiry on the role of the DNBP herbicide and the effectiveness of the soil fungicide PCNB in controlling stem rot.

The DNBP herbicide was used in experiment "B" exactly as it was in experiment "A". Cultivation also was the same as in experiment "A". PCNB was applied as a drench at the rate of 12 pounds of active substance per acre 10 weeks after planting.

Dusting in experiment "B" was exactly as in experiment "A".

During the growing seasons experiments were checked each week and infected plants were tagged and counted as soon as infection became apparent.

The peanuts were planted in early May. The Dixie Spanish variety was dug 120 days, and the Virginia Bunch variety 46-2 145 days, after planting. A one-row digger was used. In 1956 the Virginia Bunch variety 46-2 pods left in the soil at digging time were scratched out by hand from one of the 50-foot middle rows of each subplot. Three weeks after the Dixie Spanish variety was dug the peanut seedlings that emerged were counted in the middle pair of rows of

each plot. These seedlings resulted from sprouting of seeds in pods left in the soil and the sprout counts indicated the relative number of pods left in the soil at digging.

Peanuts from all experiments were cured in stacks. They were picked in mid-November with a standard farm-type picker and yields were recorded by subplots and by paired rows within each subplot. Samples for grading for size and damage were taken when the peanuts were picked.

RESULTS AND DISCUSSION

The results are presented in Tables 1 to 4 and Figure 1.

Tables 1 and 2 show a consistently higher yield and a lower incidence of infection for non-dirting weed control as compared with dirting weed control. These results attest to the effectiveness of non-dirting cultivation as a control measure for stem rot of peanuts. It is evident in Tables 3 and 4 that some part of this control was due to reduction in amount of peg rot, for there were fewer pods left in the soil when non-dirting weed control was used than when dirting weed control was used.

Boyle and Hammons (3) in referring to "conventional culture", which approximates the "dirting" treatment of this study, stated: "Besides checking the full development of the plant, this conventional method of planting and cultivation favors the development of stem blight." The Virginia study did not attempt to distinguish the effects of the cultural practices on development of peanut plants and on stem rot. Until a method is developed for making such a distinction in the taking of data it seems necessary to attribute the increase in yield for the non-dirting treatments shown in Tables 1 and 2 almost entirely to control of stem rot. It is felt that the differences in infection between non-dirting and dirting treatments as shown in the Tables are sufficiently great to support this contention.

In actual practice the non-dirting treatment would be impractical without the use of a herbicide. However, some differences that might be attributed to the fungicidal action of the DNBP herbicide are evident in results of treatments, 5, 6, 7, and 8, in Table 2. These differences are confusing, however, for the herbicide superimposed upon the non-dirting culture (treatments 5 and 6) had no significant effect upon infection but apparently did influence yield, while herbicide superimposed upon the dirting culture (treatments 7 and 8) had no significant effect upon yield but did influence infection.

Undoubtedly Table 1 shows that deep covering of surface litter in an infested field markedly reduced stem rot and increased yields in Virginia Bunch 46-2 peanuts in 1956. Table 3 indicates that this treatment tended to reduce peg rot and definitely reduced pod rot.

Peanut trash was added to plots in the experiment on Dixie Spanish peanuts in an effort to determine whether additional raw organic matter in the surface soil would result in increased incidence of stem rot. The differences between infection and yield for no added trash as compared with added trash shown in Table 2 are not statistically significant, but they do show that the added raw organic matter did not have the effect of increasing the amount of stem rot.

There are circumstances under which control of stem rot by applying fungicides to the soil might be the only feasible control. Cooper (7) in 1956 reported that of several fungicides which he applied to the soil three were somewhat effective in controlling stem rot. PCNB is the one of these three which now gives most promise. As shown in Figure 1 PCNB was applied only after it was certain that the subplots to which it was to be applied were as severely infested with Sclerotium rolfsii as were the corresponding control subplots. This figure also shows that the development of stem rot in the PCNB-treated subplots was retarded considerably after the PCNB drench was applied. The data in Table 2 show that some control of stem rot may be attributed to the fungicidal action of the PCNB. Under the conditions of this experiment PCNB was not nearly as effective in controlling stem rot as was the non-dirting without herbicide treatment. However, the PCNB-treated subplots had as many plants infected with S. rolfsii when PCNB was applied as the non-dirting without herbicide subplots had when the experiment was dug.

Quality data obtained for 2 years with Virginia Bunch 46-2 peanuts and for 1 year with Dixie Spanish peanuts present a confusing picture of the effects of the various practices on quality of the harvested peanuts. The data in Table 4, however, suggest that the PCNB drench tended to reduce peg rot, pod rot, and kernel damage in Dixie Spanish peanuts in 1956.

Table 1. Prevalence of Sclerotium rolfsii infection on Virginia Bunch 46-2 peanuts, and yields, 1955 and 1956, Holland, Virginia.

Treatment	Stand infected		Yield per acre		Yield increase	
	1955	1956	1955	1956	1955	1956
	Percent	Percent	Pounds	Pounds	Percent	Percent
1. Deep covering -- non-dirting	0.8	4.8	2924	4142	8.5	78.4
2. Deep covering -- dirting	5.1	20.0	2694	3247	base	39.9
3. Shallow covering -- non-dirting	0.8	10.1	3134	3698	16.3	59.3
4. Shallow covering -- dirting	7.8	39.2	2729	2322	1.2	base
Main plots:						
Deep covering	3.0	12.4	2790	3695**	base	22.8
Shallow covering	4.3	24.6**	2931	3010	5.0	base
Subplots:						
Non-dirting	0.8	7.4	3029**	3920**	11.7	40.8
Dirting	6.5**	29.6**	2711	2784	base	base
Replication[a]						
Subplots x main plots		*		*		

**Significant at the 1% level.
*Significant at the 5% level.
[a] In no instance was replication significant at the 5% level.

Table 2. Prevalence of Sclerotium rolfsii infection on Dixie Spanish peanuts, and yields, 1956, Holland, Virginia.

Treatment	Stand infected	Yield per acre	Yield increase
	Percent	Pounds	Percent
5. Non-dirting with herbicide	6.5[a]	2230[a]	80.4
6. Non-dirting without herbicide	8.0[a]	1941[b]	57.1
7. Dirting with herbicide	17.7[b]	1382[c,d]	11.8
8. Dirting without herbicide	27.5[c]	1236[d]	base
9. Dirting without herbicide but with PCNB[e]	17.9	1525[c]	23.4
Main plots:			
Added trash	10.5	1717	13.5
No added trash	17.4	1512	base
Subplots (treatments)	**	**	
Replications[f]			
Subplots x main plots[f]			

[a,b,c,d] Within a single column there is no significant difference (5% level) between values indicated by the same letter.
[e] Almost half of the infected plants were infected at time PCNB was applied.
[f] In no instance was either replication or subplots x main plots significant at the 5% level.
**Significant at the 1% level.

Table 3. Virginia Bunch 46-2 peanuts left in the soil at digging, 1956, Holland, Virginia.

Treatment	Total nuts per acre Pounds	Decayed nuts[a] per acre Pounds
1. Deep covering — non-dirting	238	74
2. Deep covering -- dirting	773	287
3. Shallow covering — non-dirting	826	542
4. Shallow covering — dirting	1210	735
Main plots:		
Deep covering	505	180
Shallow covering	1018*	638**
Subplots:		
Non-dirting	532	303
Dirting	991**	511**
Replications		**
Subplots x main plots[b]		

[a] The decayed nuts were counted and weights were determined by a factor derived from weight of sound nuts.
[b] In no instance was subplots x main plots significant at the 5% level.
**Significant at the 1% level.
*Significant at the 5% level.

Table 4. Dixie Spanish peanuts left in the soil at digging, percent damage, and percent of pods rotted, 1956, Holland, Virginia

Treatment	Sprouts per 400 feet of row Number	Damaged kernels Percent	Pods rotted in hand-picked sample Percent
5. Non-dirting with herbicide	275[a]	1.2[a,b]	6.2[b]
6. Non-dirting without herbicide	639[b]	1.6[b]	3.4[a]
7. Dirting with herbicide	859[b]	1.6[b]	4.9[a,b]
8. Dirting without herbicide	829[b]	1.5[b]	5.6[b]
9. Dirting without herbicide but with PCNB	294[a]	0.4[a]	3.4[a]
Main plots:			
Added trash	1731**	1.3	5.0
No added trash	871	1.3	4.4
Subplots (treatments)	**	**	**
Replications	**		
Subplots x main plots	**	*	

[a,b] Within a column there is no significant difference (5% level) between values followed by the same letter.
** Significant at the 1% level.
* Significant at the 5% level.

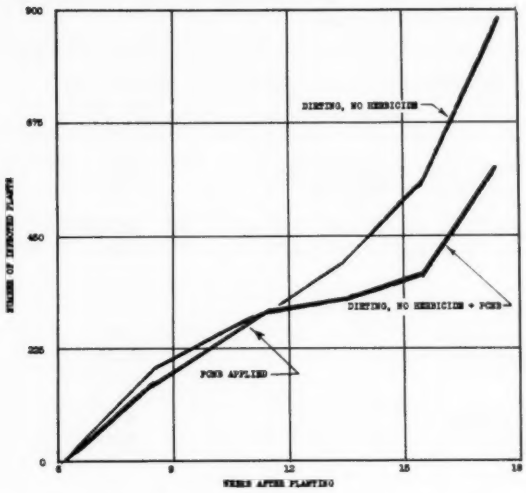

FIGURE 1. Sclerotium rolfsii infection of Dixie Spanish peanuts in the plots drenched with PCNB and in plots not drenched.

Literature Cited

1. BOYLE, L. W. 1952. Factors to be integrated in control of southern blight on peanuts. (Abstr.) Phytopathology 42: 282.
2. BOYLE, L. W. 1956. Fundamental concepts in the development of control measures for southern blight and root rot of peanuts. Plant Dis. Reptr. 40: 661-665.
3. BOYLE, L. W., and R. O. HAMMONS. 1956. Cultural practices with respect to peanut yields and control of southern blight and root rot. Georgia Expt. Sta. Mimeo. Series N. S. Bul. 31.
4. CAMPBELL, L. 1956. Control of plant disease by soil surface treatment. (Abstr.) Phytopathology 46: 635.
5. CHAPPELL, W. E., and L. I. MILLER. 1956. The effect of certain herbicides on plant pathogens. Plant Dis. Reptr. 40: 52-56.
6. COOPER, W. E. 1955. Southern stem rot. (In Perry, A. et al. "Peanut production guide for North Carolina farmers."). North Carolina Agr. Ext. Serv. Ext. Circ. 257. (Rev.)
7. COOPER, W. E. 1956. Chemical control of Sclerotium rolfsii in peanuts. (Abstr.) Phytopathology 46: 9.
8. COOPER, W. E. 1956. Progress made in stem rot control. Virginia-Carolina Peanut News. Vol. 2, No. 3. Pages 4, 6. July, 1956.
9. GARREN, K. H. 1947. Peanut disease investigations. Georgia Agr. Expt. Sta. 59th Ann. Rept., page 79.
10. GARREN, K. H. 1948. Peanut disease investigations. Georgia Agr. Expt. Sta. 60th Ann. Rept., page 64.
11. GARREN, K. H. 1957. Efficacy of certain cultural practices as control measures for stem rot of peanuts. (Abstr.) Phytopathology 47: (in press).
12. HIGGINS, B. B. 1927. Physiology and parasitism of Sclerotium rolfsii. Phytopathology 17: 417-448.
13. McCLINTOCK, J. A. 1917. Peanut wilt caused by Sclerotium rolfsii. Jour. Agr. Res. 8: 441-448.

14. McCLINTOCK, J. A. 1918. Further evidence relative to the varietal resistance of peanuts to Sclerotium rolfsii. Science (N. S.) 47: 72-73.
15. REYES, G. M. 1937. Sclerotium wilt of peanuts, with special reference to varietal resistance. Philippine Journ. Agr. 7: 245-287.
16. SING, B. and S. C. MATHUR. 1953. Sclerotial root rot disease of groundnuts in Uttar Pradesh. Current Sci. 22: 214-215.
17. TAUBENHAUS, J. J. 1919. Recent studies on Sclerotium rolfsii. Jour. Agr. Res. 18: 127-139.
18. TISDALE, W. B. 1921. Two sclerotial diseases of rice. Jour. Agr. Res. 21: 649-658.
19. WOLF, F. A. 1920. Leaf spots and some fruit rots of peanuts. Alabama Col. Sta. Bul. 180: 127-140.

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EFFECT OF SEVERAL ANTIBIOTICS AND OTHER ORGANIC CHEMICALS ON ISOLATES OF FUNGI WHICH CAUSE BEAN ROOT ROT[1, 2]

Arlen D. Davison and John R. Vaughn[3]

Bean root rot has become increasingly prevalent in the Rocky Mountain Region. Research at the University of Wyoming was undertaken in an effort to determine the value of some of the newer organic chemicals in control of the bean root rot.

The work was divided into two phases: 1) plate screening tests of compounds in the laboratory against isolates causing root rot, and 2) further screening tests of the chemicals against the root-rot disease in soil containing bean plants.

In 1919, Burkholder (2) described a dry root rot of the bean plant (Phaseolus vulgaris L.) in New York State caused by Fusarium martii Appel & Wr. var. phaseoli Burk. Others workers (3, 4) describe bean root rot caused by Fusarium solani (Mart.) Appel & Wr. var. martii (Appel & Wr.) Wr. f3 Snyder and Corticium vagum Berk. & Curt. (Pellicularia filamentosa (Pat.) Rogers).

Burke and Starr (1) reported that more than 50 chemicals and materials had been tested as seed treatments or as soil treatments. Of these, Dithane Z78, Arasan (with gypsum), New Improved Ceresan, and Spergon showed the most control. Watson (5) found that two soil fumigants, Shell CBP-55 and formaldehyde, gave excellent control of beans and pea root rot in the greenhouse and field for 2 years, the former being more effective than formaldehyde.

To date, however, the only recommended method of reducing bean root rot is to follow a rotation and try to create the most favorable growing conditions for the bean plant (6).

METHODS AND MATERIALS

Laboratory Screening Tests of Several Organic Chemicals and Antibiotics

In 1954, 20 different isolates were obtained from root rot affected beans grown at the Powell Substation in northern Wyoming. Four isolates, two of which later proved to be the most pathogenic, were chosen to use in screening tests to determine the toxicity of several antibiotics and organic chemicals. The isolates used for the testing were designated P1, P20, P22, and P24. P1, P20, and P22 were forms of Fusarium. P24 has produced no spores and has not been identified.

Vancide Z-65 (zinc salt of dimethyldithiocarbamic acid and zinc salt of 2 mercaptobenzothiazol 65%, Niagara Chemical Division); Vancide F-1665 (R. T. Vanderbilt Co., Inc.); Fungicide 275 (PCNB) (pentachloronitrobenzene 95%, Olin Mathieson Chemical Corporation); X. S. F. #3 (H. L. Woodhuysen and Associates); Crag Fungicide 974 (3-4 dimethyltetrahydro-1, 3, 5, 2H-thiadiazine-2-thione, Carbide and Carbon Chemical Company); Dithane D-14 (Nabam 19%, Rohm and Haas Company); Endomycin (Upjohn Company), were all tested at concentrations of 100, 500, and 1000 parts per million. Acti-dione (cycloheximide, Upjohn Company) was tested at 5, 50, and 100 ppm, and Ilotycin (Eli Lilly Company) at 5, 50, and 500 ppm. These compounds were supplied by the manufacturers.

The test plates were seeded by placing a 5-mm diameter disc cut from a 4-day-old culture of the test organism in the center of each plate. Three replications of each concentration and untreated media were tested with each of the three concentrations. The growth of each colony was measured in mm every two days and an average growth of all colonies determined for a period of 10 days.

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[1] Published with approval of the Director, Wyoming Agricultural Experiment Station as Journal Paper No. 91.

[2] Part of the work done in a thesis by the senior author for partial fulfillment of the Master of Science degree, University of Wyoming.

[3] Formerly Research Assistant and Professor of Plant Pathology, Wyoming Agricultural Experiment Station, respectively.

Table 1. Summary table illustrating effect of the chemicals tested against Fusarium isolate P20 in the laboratory.

Chemical	Concentration (ppm)	Maximum growth in millimeters*
Acti-dione	5	87
	50	44
	100	23
Ilotycin	5	88
	50	90
	500	75
Endomycin	100	5
	500	5
	1000	5
Vancide F-1665	100	84
	500	31
	1000	17
Vancide Z-65	100	75
	500	31
	1000	17
X.S.F. #3	100	68
	500	68
	1000	64
Fungicide 275 (PCNB)	100	58
	500	61
	1000	59
Dithane D-14	100	76
	500	40
	1000	24
Crag Fungicide 974	100	5
	500	5
	1000	5

* On untreated medium all replicates had reached the sides of the plates (approximately 90 mm).

Table 2. Effect of several chemicals on isolate P20 in soil.

Treatment	Concentration (ppm)	Average disease rating	Variation from check
Acti-dione	250	1.9	+ .1
Endomycin	1000	.7	- 1.1**
Vancide F-1665	500	.8	- 1.0*
Vancide Z-65	500	.7	- 1.1**
Dithane D-14	1000	1.4	- .4
Crag Fungicide 974	125	.7	- 1.1**
Inoculated check		1.8	
L.S.D. 5%		.8	
L.S.D. 1%		1.1	

* Significant difference at 5% level.
** Significant difference at 1% level.
- indicates less, + more, root infection than inoculated check.

Greenhouse Screening Tests

The chemicals that inhibited the growth of the isolates in the laboratory tests were later tested in the greenhouse in soil in which beans were grown.

A 4-day-old culture of each isolate was placed in 1 liter of distilled water and mixed in a Waring Blendor. One hundred ml of this suspension was mixed with a light, sandy, sterile soil in a 6-inch clay pot. The pots were allowed to stand for 3 days before beans were planted or the various chemicals added. Two varieties of beans were used, Great Northern and Asgrow Black Valentine. Five beans were planted per pot at a depth of about 1 1/2 inches.

Two methods of applying the chemicals were tested. In the first method the desired concentrations were obtained by dissolving a specific amount of the compound in 25 ml of ethyl alcohol and adding 975 ml of distilled water to make 1 liter. Fifty ml of this solution was poured on the soil in the pots every 2 days for a total of 5 applications. In the second method, 1 day prior to planting, a specific amount of the compound was added to the soil in powder form with the exception of Dithane D-14, a liquid.

All concentrations were calculated on volume basis of the compounds. At the end of 5 weeks the individual bean plants were rated according to severity of root rot. The readings were based on a 1 to 4 scale, 1 being the least severely affected, and 4 most severely.

RESULTS AND DISCUSSION

Laboratory Plate Screening Tests

In the laboratory tests, 6 of the 9 chemicals were inhibitory to one or more of the four isolates. Ilotycin and X.S.F. #3 did not inhibit any of the four test isolates enough to warrant testing in the greenhouse. Fungicide 275 (PCNB) was effective against isolates P1, P22, and P24. However, it caused only slight inhibition of P20, which was found to be the most pathogenic of the four isolates, and for this reason, it was not tested further. Crag Fungicide 974 was the most inhibitory chemical tested in the laboratory. None of the four isolates grew when placed on medium containing this chemical at any concentration used. Endomycin was the second-best chemical tested, allowing none of the four isolates to grow more than 11 mm. Vancide F-1665, Vancide Z-65, and Dithane D-14 were effective against the four isolates when used at concentrations of 500 and 1000 ppm, but at 100 ppm had very little effect against isolate P20. Acti-dione completely inhibited isolates P1 and P22 at 50 and 100 ppm but had very little effect against isolate P24. Acti-dione suppressed the growth of isolate P20 markedly at 100 ppm.

Table 1 is a summary Table illustrating the comparative effect of the nine chemicals tested against isolate P20 in the laboratory.

Greenhouse Screening Tests

Four separate trials were conducted in the greenhouse. The first two trials tested the action of Vancide Z-65, Vancide F-1665, Endomycin, and Acti-dione against isolates P1 and P20 in the soil. The same concentrations as were tested in the plate screening tests were used in both trials. Vancide Z-65 and Vancide F-1665 were effective against isolate P20 at 1000 ppm. Endomycin reduced the amount of infection caused by isolate P20 but the reduction was not significantly different from the check. Acti-dione had very little effect on isolate P20 in the soil. Vancide Z-65, Vancide F-1665, Endomycin, and Acti-dione were all effective against isolate P1 when applied to the soil.

In the third trial Vancide Z-65, Vancide F-1665, Endomycin, and Dithane D-14 were tested at concentrations of 1000, 2000, 3000, and 4000 ppm against isolate P20. Acti-dione was tested at concentrations of 100, 1000, 2000, and 3000 ppm. The treatments with Dithane D-14 were significantly different from the check but the chemical caused mottling and stunting, and swelling of the lower stem of the bean plants. Acti-dione caused severe dwarfing and yellowing of the bean plants at the higher concentration and had no inhibitory effect on isolate P20. Endomycin, Vancide Z-65, and Vancide F-1665 at one or more of the concentrations tested reduced the amount of disease significantly below the check plants.

Results from the fourth trial indicated no significant reduction in the amount of root rot when the data for all five isolates were analyzed as a complex analysis of variance. However, by running a separate analysis on isolate P20 alone it was found that Endomycin, Vancide F-1665, Vancide Z-65, and Crag Fungicide 974 significantly reduced root-rot infection by isolate

P20 at the 5 percent level. It is unusual that a chemical which is effective against one isolate should not be effective against related species, and it is interesting to note that the less pathogenic isolates were in this case most resistant.

Table 2 is a summary table of greenhouse trials illustrating the effect of several chemicals on isolate P20 in the soil.

SUMMARY AND DISCUSSION

Vancide Z-65 was the most effective chemical tested against isolates P20 and P1. Vancide F-1665 and Endomycin were about equally effective against isolates P20 and P1. Vancide Z-65 and Vancide F-1665 consistently reduced the amount of root rot. Endomycin reduced the amount of root rot in three out of four trials. Endomycin, Vancide F-1665, Vancide Z-65, and Crag Fungicide 974 were all effective against isolate P20 but did not significantly reduce the amount of root rot caused by several other isolates of Fusarium. Although none of the chemical treatments resulted in the degree of control desired, several were promising enough to warrant further testing, particularly Vancide Z-65 and Crag Fungicide 974.

Literature Cited

1. BURKE, D. W., and G. H. STARR. 1950. Studies on bean root rot in Northern Wyoming. (Abst.). Jour. Colo.-Wyo. Acad. Sci. 4(2): 58-59.
2. BURKHOLDER, W. H. 1919. The dry root rot of bean. Cornell Univ. Agr. Exp. Sta. Memoir No. 26: 999-1033.
3. BUTLER, EDWIN J. and S. G. D. JONES. 1949. Root rot of French bean. Plant Pathology. Macmillan and Co., Ltd. London: 607-608.
4. MURPHY, D. M. 1940. Bean improvement and bean diseases in Idaho. Idaho Agr. Exp. Sta. Bul. 238: 1-22.
5. WATSON, R. D. 1953. Control of root rot by row application of soil fumigants. (Abst.). Phytopathology: 43: 590.
6. ZAUMEYER, W. F. and H. REX THOMAS. 1953. Field diseases of beans and lima beans. In Plant Diseases, the Yearbook of Agriculture. United States Department of Agriculture: 393-400.

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COMPARISON OF THE EFFECT OF SOME ANTIBIOTICS, ANTIFUNGAL SUBSTANCES, AND PHENYL CARBAMATES ON THE GROWTH OF TWO VASCULAR PARASITES IN VITRO

I. R. Schneider[1]

Summary

The imperfect stages of two vascular parasites Ceratostomella ulmi Buism.[2] and Endoconidiophora fagacearum Bretz[2], were grown in laboratory culture. Antibiotics, antifungal substances, and phenyl carbamates, were added to media, and the effect of the added substances on diameter growth of the two parasites was compared. Most substances tested inhibited both organisms by approximately the same magnitude. Exceptions to the usual uniform inhibition resulted with five antibiotics and one carbamate: endomycin, fradicin, pleocidin, gramicidin S, cycloheximide, and propynyl N-phenyl carbamate. Five times the concentration of cycloheximide that completely prevented the growth of E. fagacearum, the oak wilt fungus, did not significantly inhibit the growth of C. ulmi, the Dutch elm disease fungus. Pleocidin was also more inhibitory to the oak wilt fungus, while gramicidin S, endomycin, fradicin, and propynyl N-phenyl carbamate were more inhibitory to the Dutch elm disease fungus.

The compounds most active against both organisms were: endomycin, fradicin, fungichromin, ascosin, candicidin, several derivatives of 2-pyridinethiol-1-oxide (Olin Mathieson numbers 1562, 1564, 1456, and 1484), mycostatin, isopropyl N-chlorophenylcarbamate, alpha-carboxyethyl N-3-chlorophenylcarbamate, propynyl N-phenylcarbamate, 2-propynyl N-phenylcarbamate, ethyl N-phenylcarbamate, isopropyl N-(3-chlorophenyl)-carbamate and 2-(1-chloropropyl) N-phenylcarbamate.

Both C. ulmi and E. fagacearum were exposed for a period of 24 hours, 3 days, or 7 days to each of the more active compounds at concentrations above that required for complete inhibition. The fungi were then transferred from a medium containing an added compound to the same medium free of the substance. Under these conditions, growth of each fungus was visible within one week after exposure to each antibiotic (except cycloheximide at 25 times the minimum concentration which prevented growth of E. fagacearum), indicating fungistatic and not fungicidal action. Some of the 2-pyridinethiol-1-oxide derivatives were fungicidal as were two phenyl carbamates.

INTRODUCTION

The causal agents of oak wilt and the Dutch elm disease are vascular parasites, capable of causing the rapid death of their respective hosts. The growth of each organism in culture has been studied individually from the standpoint of nutritional sources (2, 3, 6, 12, 14). Both organisms appear able to utilize ammonium or organic nitrogen as nitrogen sources. Various carbon sources are reported suitable for both organisms. Lactose and galactose were reported good carbon sources for Ceratostomella ulmi Buism. (12). The suitability of galactose as a source of carbon differs somewhat with the investigator (2, 3), but lactose was a poor carbon source for Endoconidiophora fagacearum Bretz (2, 3, 6). A striking difference exists in a vitamin requirement. C. ulmi is deficient for pyridoxine (9, 10), but this vitamin does not appear necessary for growth of the oak wilt fungus (3, 6). Other differences are in-

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[2] [Ceratocystis ulmi (Buism.) C. Moreau, and C. fagacearum (Bretz) Hunt (Hunt, John. Taxonomy of the genus Ceratocystis. Lloydia 19: 1-58. 1956) -- Ed. Note.]

dicated in that C. ulmi grows well on Difco potato dextrose agar, but E. fagacearum grows poorly on this medium[3].

It was felt that a comparison of the effects of compounds[4] on the growth of C. ulmi and E. fagacearum would reveal new similarities and differences useful in a chemotherapeutic program involving these two parasites. The author does not wish to imply, however, that a therapeutant to be successful in vivo must necessarily be toxic to the pathogen in vitro.

MATERIALS AND METHODS

In the standardized procedure followed, a 1-ml aliquot of the compound to be tested was incorporated in 19 ml of warm melted agar medium per Petri dish. Uniform distribution of the chemical was obtained by agitation. Antibiotics and antifungal substances were tested either at 8, 40 and 200 ppm, or at 4, 20, and 100 ppm. All phenylcarbamates were tested at 20, 100, and 500 ppm. Each concentration was tested in triplicate.

Agar disks, each 5 mm in diameter, were cut with an alcohol-flamed cork borer from the periphery of the colony of the fungus to be tested. Each disk was transferred, inverted, to the center of a Petri dish which contained Difco PDA in the case of C. ulmi and bran extract agar[5] in the case of E. fagacearum. Each agar disk included mycelium and spores of the fungus to be tested. Controls consisted of the appropriate medium without a test chemical incorporated to which a 5 mm disk of either fungus was transferred. At the end of 7 days incubation at 24°C, diameter measurements were taken in two directions, and averaged for each triplicate determination. The percentage inhibition of growth attributable to each chemical was calculated from the measurements.

RESULTS

Most substances tested inhibited both organisms by approximately the same magnitude. Among the antibiotics studied, five resulted in a significantly greater inhibition of one organism than of the other (Table 1). These were endomycin, fradicin, pleocidin, gramicidin S, and cycloheximide. Cycloheximide resulted in the largest difference in inhibition. At 8 ppm it prevented growth of the oak wilt fungus, yet at 200 ppm it did not cause significant inhibition of the Dutch elm disease fungus.

The phenylcarbamates differed considerably in activity. Inhibition of the two fungi ranged from 0 to 100 percent depending on the derivative tested. However, with the exception of propynyl N-phenylcarbamate, each phenylcarbamate inhibited both fungi approximately the same amount. Propynyl N-phenylcarbamate prevented the growth of C. ulmi at 100 ppm, while 500 ppm were required to prevent growth of E. fagacearum (Table 2).

Antibiotics and antifungal substances with relatively high activity in the inhibition of diameter growth of both C. ulmi and E. fagacearum are are shown in Table 1. Derivatives of 2-pyridinethiol-1-oxide (Olin Mathieson 1562, 1564, and 1456) prevented growth of E. fagacearum at only 4 ppm. Alkaline fradicin at 8 ppm prevented growth of the Dutch elm disease fungus; and 20 ppm fungichromin was equally effective against each fungus.

Phenylcarbamates with relatively high activity are shown in Table 2. Propynyl N-phenylcarbamate was the only phenylcarbamate studied that prevented the growth of either fungus at 100 ppm. Growth of the oak wilt fungus was prevented by a concentration of 500 ppm of propynyl N-phenylcarbamate or 2-(1-chloropropyl) N-phenylcarbamate. The same concentra-

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[3]Schneider, I. R., unpublished data.

[4]The author wishes to express his appreciation for compounds made available to him by the following people:

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Dr. P. H. Hidy, Commercial Solvents Corporation

[5]Himelick, E. B., Natural History Survey, Unpublished Data

Table 1. Antibiotics and antifungal compounds causing relatively high inhibition of either or both Ceratostomella ulmi and Endoconidiophora fagacearum (50 percent or more inhibition).

Antibiotic or antifungal compound	C. ulmi		E. fagacearum	
	Concentration in ppm	Percent inhibition	Concentration in ppm	Percent inhibition
Cycloheximide	200	9	8	100
Pleocidin	100	34	100	74
Gramicidin S	100	72	100	26
Endomycin	8	88	8	57
Fradicin (alkaline)	8	100	8	64
Fungichromin	20	100	20	100
Ascosin	100	100	100	75
Candicidin	20	68	20	72
O.M. 1562[a]	4	91	4	100
O.M. 1564[b]	4	91	4	100
O.M. 1456[c]	4	86	4	100
O.M. 1484[d]	4	68	4	70
Mycostatin	20	88	20	95
Thiolutin	20[e]	73	100	100

[a]Cu salt of 2-pyridinethiol-1-oxide

[b]Mn salt of 2-pyridinethiol-1-oxide

[c]Disulfide derivative of 2-pyridinethiol-1-oxide

[d]Na salt of 2-pyridinethiol-1-oxide

[e]No data available at 100 ppm.

tions of 2-propynyl N-phenylcarbamate, and ethyl N-phenylcarbamate prevented growth of both fungi.

Antibiotics which caused 50 percent inhibition or less of each organism at the highest concentration tested were the following: streptomycin sulfate, polymixin B sulfate, tetracycline, isonicotinic hydrazide, terramycin hydrochloride, bacitracin, penicillin G (potassium salt), noformicin, streptothricin sulfate (inhibited C. ulmi 52 percent at 100 ppm), lupulone, subtilin, quercitin, d-usnic acid, PA-96, catenulin, and carbomycin.

The phenylcarbamates which caused less than 75 percent inhibition of both fungi at 500 ppm were: alpha-carbobenzoxyethyl N-phenylcarbamate, alpha-carbobutoxyethyl N-3-methylphenylcarbamate, alpha-carbo-(2-chloroethoxy) ethyl N-phenylcarbamate, beta-cyanoethyl N-phenylcarbamate, alpha-carbo-(2, 4-dichlorophenoxy)-ethoxyethyl N-phenylcarbamate, alpha-carbobutoxyethyl N-3-chlorophenylcarbamate, alpha-carbobutoxyethyl N-methyl-N-phenylcarbamate, alpha-carbobutoxyethyl N-3-cyanophenylcarbamate, alpha-carbododecoxyethyl N-phenylcarbamate, alpha-carboxyethyl N-phenylcarbamate, alpha-carbobutoxyethyl N-3-nitrophenylcarbamate, alpha-carboxycyclohexoxyethyl N-phenylcarbamate, alpha-carbotutoxyethyl N-phenylcarbamate, and isopropyl N-phenylcarbamate.

Most of the compounds studied were not fungicidal, even at concentrations above those capable of preventing growth (Table 3). Of those compounds fungicidal to at least one fungus, several were more effective in preventing the growth of the Dutch elm disease fungus after it

Table 2. Phenyl carbamates which caused more than 75 percent inhibition of both C. ulmi and E. fagacearum.

Phenyl carbamate	C. ulmi		E. fagacearum	
	Concentration in ppm	Percent inhibition	Concentration in ppm	Percent inhibition
Isopropyl N-chloro-PC[a]	500	88	500	91
Alpha-carboxyethyl N-3-chloro-PC	500	79	500	77
Propynyl N-PC	100	100	100	37
Propynyl N-PC	500	100	500	100
2-Propynyl N-PC	500	100	500	100
Ethyl N-PC	500	100	500	100
Isopropyl N-(3-chloro-phenyl)-C[b]	500	83	500	88
2-(1-chloropropyl) N-PC	500	92	500	100

[a]PC: phenyl carbamate

[b]C : carbamate

was removed from the chemical. These were Olin Mathieson Nos. 1456 at 20 and 100 ppm, 1564 at 4 and 20 ppm, 2-propynyl N-phenylcarbamate, and ethyl N-phenylcarbamate at 1000 ppm. At 20 ppm Olin Mathieson No. 1484 appeared fungicidal to the oak wilt fungus but not to the Dutch elm disease fungus. After 7 days exposure to Olin Mathieson No. 1562 at 100 ppm both C. ulmi and E. fagacearum were still viable. In contrast, Olin Mathieson No. 1564 at 100 ppm, with 24 hours contact, was fungicidal to both fungi.

The lack of effect of 200 ppm cycloheximide on growth of the Dutch elm disease fungus was pointed out above. The concentration of 40 ppm cycloheximide was fungistatic, but not fungicidal to the oak wilt fungus, even though one-fifth this concentration prevented growth of the fungus. A concentration of 200 ppm was fungicidal after 3 days exposure, but not after 24 hours exposure.

DISCUSSION

The phenylcarbamates are plant growth regulators rather than antifungal agents. A wide range of antifungal activity was exhibited among the few members of the group that were tested. Derivatives of propynyl N-phenylcarbamate, 2-propynyl N-phenylcarbamate, and ethyl N-phenylcarbamate at 500 ppm prevented the growth of both C. ulmi and E. fagacearum while alpha-carbododecoxyethyl N-phenylcarbamate and alpha carboxyethyl N-phenylcarbamate at the same concentration caused no significant inhibition of either fungus.

The resistance of C. ulmi to high concentrations of cycloheximide has been noted previously (7, 8, 11). The resistance of the Dutch elm disease fungus to high concentrations of cycloheximide is interesting in view of the long list of fungi to which this antibiotic is toxic (1, 4, 5, 13). The variability in response shown by C. ulmi and E. fagacearum to some of the other compounds emphasizes the need to study even closely related pathogens individually.

Most compounds tested were not fungicidal. Those that were fungicidal usually required high concentrations and/or long periods during which the fungus had to be exposed to the chemical for its lethal action. This requirement is an important consideration in the attempt to destroy a parasite within its host. A chemical acting primarily on the pathogen, but not lethal to it without long contact at high concentrations, will not be likely to destroy the pathogen without injury to the host.

Table 3. The minimum time required for fungicidal action of compounds capable of preventing growth of C. ulmi and E. fagacearum.

Compound	Concentration ppm	Minimum time	
		C. ulmi	E. fagacearum
O.M. 1456	20	24 hrs.	3 days
	100	24 hrs.	3 days
O.M. 1484	20	a	7 days
	100	24 hrs.	24 hrs.
O.M. 1564	4	7 days	a
	20	3 days	a
	100	24 hrs.	24 hrs.
O.M. 1562	100,	a	a
Cycloheximide	40	b	a
	200	b	3 days
Fradicin	40	a	a
	200	a	a
Fungichromin	40	a	a
	200	a	a
Candicidin	20	a	a
	100	a	a
Mycostatin	100	a	a
Propynyl N-PC[c]	1000	a	a
2-Propynyl N-PC	1000	7 days	a
Ethyl N-PC	1000	7 days	a
2-(1-chloropropyl) N-PC	1000	a	a

[a]Not fungicidal at concentration indicated in 7 days or less exposure.
[b]Not fungistatic.
[c]PC: phenyl carbamate

Literature Cited

1. ANDERSON, H. W, and DAVID GOTTLIEB. 1952. Plant disease control with antibiotics. Economic Botany 6: 294-308.
2. BARNETT, H. L., and V. G. LILLY. 1952. Physiological factors affecting growth and sporulation of Chalara quercina in culture. (Abst.). Phytopathology 42: 2.
3. BECKMAN, C. H., J. E. KUNTZ, and A. J. RIKER. 1953. The growth of the oak wilt fungus with various vitamins and carbon and nitrogen sources. Phytopathology 43: 441-447.
4. FELBER, IRMA M., and C. L. HAMNER. 1948. Control of mildew on bean plants by means of an antibiotic. Bot. Gaz. 110: 324-325.
5. FORD, JARED H., and WILLIAM KLOMPARENS. 1956. Cycloheximide (Acti-dione) and its agricultural uses. Mimeographed.
6. HOFFMAN, P. F. 1954. Physiology of Endoconidiophora fagacearum Bretz, I. Factors influencing growth and toxin production. Iowa State College Journal of Science 29: 27-38.
7. HOLMES, F. W. 1955. Field and culture tests of antibiotics against Graphium ulmi. (Abst.). Phytopathology 45: 185.
8. HOLMES, F. W. 1954. The Dutch elm disease as investigated by the use of tissue cultures, antibiotics, and pectic enzymes. Thesis, Cornell University.
9. ROBBINS, WM. J., and ROBERTA MA. 1942. Vitamin deficiencies of Ceratostomella. Bull. Torrey Bot. Club 69: 184-203.
10. ROBBINS, WM. J., and ROBERTA MA. 1942. Specificity of pyridoxine for Ceratostomella ulmi. Bull. Torrey Bot. Club 69: 342-352.
11. SCHNEIDER, I. R. 1956. A selective medium for the routine isolation of Graphium ulmi Schwarz. Plant Dis. Reptr. 40: 816-821.
12. STEINBERG, ROBERT A. 1939. Growth of fungi in synthetic nutrient solution. Botanical Review 5: 327-350.
13. WHIFFEN, ALMA J. 1950. The activity in vitro of cycloheximide (Acti-dione) against fungi pathogenic to plants. Mycologia 42: 253-258.
14. WHITE, IRENE, and F. T. WOLF. 1952. Preliminary report on some nutritional requirements of Chalara quercina. (Abst.) Phytopathology 42: 288.

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CHEMICALLY INDUCED RESISTANCE TO STEM RUST OF WHEAT BY DERIVATIVES OF ACTI-DIONE[1]

R. G. Hacker and J. R. Vaughn[2]

Summary

The semicarbazone of cycloheximide, one of several new derivatives of Acti-dione, induces pre-infection resistance to black stem rust (Puccinia graminis tritici) in the spring wheat variety Onas. This chemical resistance persists for the life of the plant after treatment at the boot stage. A sister compound, the acetate of cycloheximide, behaves similarly, effecting good control of rust, but does not impart complete resistance. Both materials are only slightly phytotoxic at effective concentrations and have no adverse effect on yield or germination.

INTRODUCTION

Acti-dione has been tested against wheat stem rust (Puccinia graminis tritici) with varying degrees of success by several investigators. Livingston (1) believed it had "some value" both as an eradicant of rust and in imparting immunity to wheat plants. Silverman and Hart (2) found that Acti-dione produced partial control of rust at 20 to 25 ppm but that it was definitely phytotoxic at higher concentrations. Wallen (3) reported that this material gave almost complete control in field plots sprayed every 10 days with a 500 ppm solution at the rate of 110 cc per rod row. His data showed yields which were markedly higher than controls in spite of phytotoxic effects of the antibiotic. Excepting Wallen, it has been the consensus of workers using Acti-dione that the narrow range between its effective and phytotoxic levels seriously limits its practical usefulness as a control for stem rust.

New derivatives of Acti-dione[3] released for testing in the spring of 1956 have now produced what can be described best as a long-term pre-infection resistance to rust in spring wheat, without the severe phytotoxic effects of the parent compound.

One of these analogues, the semicarbazone of cycloheximide, induced total resistance to rust infection for the balance of the life of the plant after being applied at the early boot stage and again at full flower. A sister compound, the acetate of cycloheximide, afforded good protection of like nature. The lowest effective concentration was 100 ppm for the semicarbazone and 200 ppm for the acetate. Phytotoxicity was very slight at these concentrations, yields were higher than checks, though not significantly, and germination of seed from treated plots was normal.

I. GREENHOUSE TESTS

Procedure

Initial testing of the new Acti-dione derivatives was done in the greenhouse on 21-day-old Onas seedlings and on older plants, in flower, which were already heavily infected with rust. Treatments were designed to determine 1) effective concentrations and phytotoxicity levels, 2) possible inhibitory effect of applications made before inoculation and infection, 3) effect of applications made after infection, at the time of first pustule formation, and 4) the effect of these materials on well-established rust infections on older plants.

Concentrations used were 10, 50, and 100 ppm in aqueous solutions which were applied as sprays. The pre-infection applications were made 72 hours before inoculation and incubation.

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[1]Published with approval of the Director, Wyoming Agricultural Experiment Station, as Journal Paper No. 92.

[2]Graduate Research Assistant and Professor of Plant Pathology, Department of Agronomy, University of Wyoming.

[3]Materials supplied by the Upjohn Company, Kalamazoo, Michigan.

Results

Table 1 summarizes the results of these trials. Plants receiving pre-infection applications of the oxime at 100 ppm were entirely free of infection 2 weeks after inoculation. Those treated with the semicarbazone at 100 ppm developed suppressed, resistant-type pustules. All checks were heavily infected. Both materials were moderately effective as pre-infection treatments at 50 ppm, but ineffective at 10 ppm. Phytotoxicity at 100 ppm was very slight. Post-infection applications failed to stop rust development on either the seedlings or the older plants.

II. FIELD TESTS

Procedure

The field experiment was arranged in a randomized block of rod-row plots, replicated 4 times. The plots were 10 feet long and 4 rows wide with a row spacing of 14 inches. Data were taken from the center 8 feet of the middle 2 rows. The materials were applied as pre-infection and post-infection sprays in concentrations of 50, 100, and 200 ppm, at a rate of 160 cc of solution per plot. The equipment used was a 5-gallon John Bean Spray-Pal gasoline-powered pump equipped with a single Tee-Jet nozzle.

The test area is in a location where climatic conditions are far different than in other wheat-growing districts. The University of Wyoming Agronomy Farm, where this work was done, is situated in the high Laramie Basin at about 7,300 feet above sea level. The growing season is highly favorable, but short, having 90 days frost-free. Nights are cool and the average relative humidity in summer is low.

Cereal crops grown here under irrigation yield very well. Onas is the highest-yielding spring wheat variety at this altitude and in favorable years it produces 80 to 100 bushels per acre. However, being highly susceptible to rust, this yield is frequently reduced by more than 50 percent and in some years by as much as 70 percent by the rust. In spite of the seemingly adverse climatic conditions, moderate to heavy cereal-rust infection can be expected almost certainly each year.

The spore showers which produce the wheat stem rust epiphytotics usually occur around late July or early August.

Plots for this experiment were seeded in late May in order to have succulent growth in the stands when rust infection occurred. In anticipation of spore showers, the first pre-infection sprays were put on July 23, when the plants were in early boot. However, when infection failed to appear by August 3, in the check, the same plots were treated a second time. In the following two weeks all plots were artificially inoculated twice with spore-talc suspension. Rust appeared shortly afterward and became epidemic by September 10, at which time final rust readings were taken. Conditions were further complicated by a light frost on August 27, which nipped upper leaves, and a killing frost on September 5, which terminated growth entirely.

Results

Phytotoxicity (Table 2) -- The readings given represent visual estimates of the apparent effect of the antibiotics seven days after the initial application. The rating of "slight" was given where leaves of some plants within a treatment exhibited a slight marginal scalding. In no case was there any general tissue damage by either compound. In all plots, heads were erect and normal.

Rust Readings (Table 3) -- All plots receiving the semicarbazone analogue at 200 ppm were completely free of rust. A thorough row-by-row inspection of all replicates receiving this treatment did not reveal a single pustule. Check plots carried infection up to 65 percent severity (average plot condition); on individual plants the rating was 100 percent. The average severity for all check plots in all replicates was 20 percent and the average prevalence was 100 percent.

The semicarbazone treatment at 100 ppm was almost as effective as the 200 ppm dosage. Two of the replicates had no infection, but a few scattered pustules in the other two plots made it necessary to rate the average condition for the treatment as "trace" in all categories. The 50 ppm treatment gave fair control but was less effective.

The acetate treatments, which showed so much promise in the greenhouse tests, lagged behind the results achieved by the semicarbazone analogue in the field. Even so, this material gave very good control at the 200 ppm concentration, which would indicate that it is worthy of further testing.

The readings given in Table 3 are estimates based on the Cobb Scale and represent averages for all the replicates in each treatment. These are stem readings only, taken from the basal one-half of the plants. Leaf readings were not possible, due to the early frost damage mentioned previously. In this connection, it is of interest that the lower stems remained green through the two periods of frost and that the progress of rust development apparently was not slowed.

Rod-row Yields (Table 4) -- Average yields given in Table 4 show a slight increase for all treatments over the control, except from those plots receiving the acetate at 200 ppm. An analysis of variance indicated that none of these differences was significant. The fact that there was no reduction in yield emphasizes the safety of these materials.

Germination Counts (Table 5) -- Seed harvested from all the treated plots had high, normal germination. Tests were run by the Wyoming State Seed Laboratory, and the blotter counts were verified by bench plantings in the greenhouse. This was done principally to make certain that the seedlings produced had normal vigor. Analysis of variance showed that the differences in germination percentages in Table 5 are not significant.

It was noted that seedlings from the 200 ppm semicarbazone treatment emerged sooner and seemed slightly more vigorous than those from the checks or other treatments. Seven days after planting, there were approximately 35 percent more seedlings in the four replicates of this treatment than in the others. A uniform-depth planting bar had been used in seeding this test and the treatments were randomized.

Tests on Second Generation Plants -- There was no carry-over of resistance in second-generation plants grown from seed harvested off the plots treated with the 200 ppm concentrations of both compounds. Inoculated seedlings developed vigorous, susceptible-type pustules typical of the variety.

DISCUSSION

The most outstanding characteristic of these new materials, aside from their apparent therapeutic potency, is their long-term effect after application. Plants treated with the 200 ppm concentration of the semicarbazone analogue were totally resistant all the way to maturity -- a span of 38 days from the last application. It is not possible to postulate at this time on the mode of action of this antibiotic, so it is sufficient to state that it appears to impart a chemical resistance, either directly or indirectly, to the establishment of the rust organism in the host tissue. The manufacturer of these compounds has established their systemic nature and has classified them as systemic anti-fungal agents. Accordingly, the control effect is presumed to occur within the plant. However, the possibility that there may also be some inhibition of spore germination by residue on the stems and leaves cannot be excluded.

Another significant point is the almost complete lack of phytotoxicity of these materials at effective concentrations. In spite of the fact that the second treatment was made when the plants were in full flower, yields and germination were not impaired. On the contrary, there was some evidence that the seed from the semicarbazone treatments germinated more vigorously than that from the non-treated plots.

It was suggested previously that results of these experiments would have to be interpreted in the light of climatic and certain other conditions under which the work was conducted. Yield data, particularly, were subject to the influence of early damaging frosts before maturity. Total yield weights for all plots were considerably lower than the normal expected yield for the variety. The question arises whether this over-all decrease would mask any increases or decreases due to the treatments. Also the influence of rust infection on yields from the checks should be considered. It is felt that it was probably negligible, since the heads were so near maturity when the rust became established. Translocation of elaborated foods no doubt had fallen to near zero on September 5, when severe frost killed all the leaves. Rust on the lower stems did not become epidemic until 5 days later.

Table 1. Degree of rust control by various concentrations of the cycloheximide analogues in preliminary greenhouse trials.

Concentrations	Semicarbazone			Acetate		
	Preinfection Spray	Postinfection Spray	Effect on established infection	Preinfection Spray	Postinfection Spray	Effect on established infection
10 ppm	0	0	0	0	0	0
50 ppm	Moderate	0	0	Moderate	0	0
100 ppm	Good (resistant type pustules)	0	0	Complete Control	0	0

Table 2. Foliar phytotoxicity of cycloheximide analogues

Concentration	Severity	
	Semicarbazone	Acetate
50 ppm	0	0
100 ppm	Trace	Trace
200 ppm	Slight	Slight

Table 3. Field rust readings using the Cobb Scale

Concentration	Semicarbazone			Acetate		
	Average Severity	Maximum Severity	Average Prevalence	Average Severity	Maximum Severity	Average Prevalence
50 ppm	Trace +	40	10	5 +	40	65
100 ppm	0 to Trace	Trace	Trace	5 -	25	40
200 ppm	0	0	0	Trace	10 +	10 +
Check	20	65	100	20	65	100

Table 4. Field test yields in bushels per acre

Treatment	Semicarbazone		Acetate	
	Yield	Percent of check *	Yield	Percent of check *
50 ppm	40.9	106	42.3	110
100 ppm	42.9	111	41.6	107
200 ppm	41.9	109	38.2	98
Check	38.6	100	38.6	100

* Apparent differences not statistically significant

Table 5. Germination percentages of seed from field tests

Treatment	Semicarbazone		Acetate	
	Blotter Tests	Bench Plants	Blotter Tests	Bench Plants
50 ppm	91	94	87	90
100 ppm	96	92	94	90
200 ppm	94	92	94	92
Check	93	92	93	92

CONCLUSIONS

1. Pre-infection applications of the semicarbazone of cycloheximide controlled black stem rust on a highly susceptible spring wheat variety; its effect persisted for a long period of time after application; and phytotoxicity was negligible since there was no adverse effect on yield or germination.
2. The acetate of cycloheximide, though not so effective in field tests, shows promise.

Literature Cited

1. HACKER, ROBERT G., and JOHN R. VAUGHN. 1957. Cycloheximide analogues cause preinfection resistance to Puccinia graminis var. tritici in spring wheat. Phytopathology 47: 14.
2. LIVINGSTON, J. E. 1953. The control of leaf and stem rust of wheat with chemotherapeutants. Phytopathology 43: 496-499.
3. SILVERMAN, W. B., and HELEN HART. 1954. Antibiotics tested for control of wheat stem rust. Phytopathology 44: 506.
4. WALLEN, V. R. 1955. Control of stem rust of wheat with antibiotics. Plant Dis. Reptr. 39: 124-127.

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DEGRADATION OF CYCLOHEXIMIDE DERIVATIVES IN PLANTS

A. J. Lemin and W. E. Magee

Abstract

The uptake and movement of cycloheximide acetate-2-C^{14} into tomato plants has been examined. It was discovered that cycloheximide acetate was absorbed by the roots and induced strongly antifungal properties in the leaves. This activity was probably not due to the acetate but, more likely, to the formation in the leaves of free cycloheximide.

The systemic activity of a number of cycloheximide[1] derivatives in plants has been reported (2, 3, 5). Work in our laboratories has shown that some cycloheximide derivatives have systemic antifungal activity on economically important parasites of tomato, wheat, apple, and cherry.

A number of experiments were performed in an attempt only to trace the uptake and degradation of one of these derivatives in the plant. Intact tomato plants (variety Bonnie Best) were allowed to stand in a solution of cycloheximide acetate in water (100 mcg/ml) until pronounced wilting of the plants was noted, usually in 6 to 8 hours. No wilting was observed when plants were allowed to stand for this period in water or in solutions of sodium acetate.

The plants which had been treated with cycloheximide acetate were harvested, solvent extracted, and the extracts examined by paper chromatography (Bush B-5 solvent system). The presence and location of activities were determined by a bio-assay technique using Saccharomyces pastorianus (6). A spot corresponding to cycloheximide acetate (Rf 0.8) and a spot (Rf ca. 0.45) corresponding to cycloheximide could be detected on the paper chromatograms by treatment of the paper with phosphomolybdic acid. The zone with Rf 0.45 (cycloheximide) inhibited the growth of S. pastorianus on plates while the zone of Rf 0.8 did not.

The observation that cycloheximide acetate produced a material with high antifungal activity when translocated in tomato plants led to a further series of experiments. Radioactive cycloheximide acetate labeled only in the carbon two position of the acetate moiety was prepared from cycloheximide and acetic anhydride-2-C-^{14} by the method of J. H. Ford and B. E. Leach (1) and the labeled derivative was then used to trace the rate of uptake through the roots of intact tomato plants. Six plants were used, the roots of each plant being placed in contact with 25 ml of a solution of 10 mg of cycloheximide acetate-2-C^{14} in water. Two plants were harvested, roots, stems, and leaves separately, at each time interval (4, 7 1/2, and 11 hours) and the tissues extracted with boiling 80% methanol.

It was found that absorption through the roots of the active compound was rapid and that it continued throughout the course of the experiment, with a corresponding decrease in the radioactivity of the remaining soak solution (Table 1). Further, although a saturation level was soon reached in the roots and stems of the plants, the leaves continued to show increased radioactivity until the close of the experiment. The highest level of radioactivity and the highest bio-activity after 11 hours were found in the second unfolded leaf from the top of the plant.

Paper chromatography of the extracts from the washed stems and roots from this and another similar experiment showed that most of the radioactivity was in a region on the paper corresponding to cycloheximide acetate. Chromatography of the leaf extracts showed radioactivity in a number of areas on the paper with very little corresponding to cycloheximide acetate. In another similar experiment employing lower levels of isotope, the intact cycloheximide acetate-2-C^{14} could be demonstrated only in the root extract. Thus, the radioactive portion was cleaved from the molecule and metabolized by the plant tissue. Most of the fractions showed higher counting rates when dried in the presence of alkali, suggesting that liberated acetic acid-2-C^{14} and perhaps $C^{14}O_2$ were being lost.

This may explain the incomplete recovery of radioactivity since gaseous metabolic products were not collected in the experiment. Bio-assay of the leaf extracts demonstrated that

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[1] Trade name "Acti-dione", The Upjohn Company, Kalamazoo, Michigan

Table 1. The incorporation of radioactive Acti-dione acetate-2-C^{14} into the tomato plant.

	Total counts per minute[a] (in two plants)		
	4 hours	7 1/2 hours	11 hours
Initial radioactivity added	7.36×10^5	7.46×10^5	7.46×10^5
Final radioactivity in solution	3.08×10^5	2.2×10^5	1.47×10^5
Alcohol extract of root	0.322×10^5	0.195×10^5	0.302×10^5
Alcohol extract of stem	0.189×10^5	0.415×10^5	0.475×10^5
Alcohol extract of leaf (total)	0.128×10^5	0.367×10^5	0.645×10^5
Total recovery of radioactivity	3.719×10^5	3.187×10^5	2.792×10^5

[a] The fractions were made alkaline with potassium hydroxide and counted as infinitely thin samples using a micro-mil thin window counter (Nuclear-Chicago) with 35% geometry.

Table 2. Antifungal activity in the tomato tissue derived from cycloheximide acetate-2-C^{14} [b].

	Equivalent total mcg activity in the extracts (two plants)		
	4 hours	7 1/2 hours	11 hours
Alcohol extract of root	102	107	168
Alcohol extract of stem	115	404	413
Alcohol extract of leaves	133	404	1023

[b] No *in vitro* activity of cycloheximide acetate using levels up to 1000 mcg/ml has been observed in this bio-assay. The equivalent total mcg was calculated using cycloheximide as a standard.

the antifungal activity (Rf 0.45) showed a rapid increase with time (Table 2).

These experiments suggest that cycloheximide acetate may be absorbed intact through the roots of the tomato plant, but that it derives its antifungal activity from free cycloheximide released in the plant.

Literature Cited

1. FORD, J. H., AND B. E. LEACH. 1948. Acti-dione, an antibiotic from Streptomyces griseus. Jour. Amer. Chem. Soc. 70: 1223.
2. HACKER, R. G., and J. R. VAUGHN. 1957. Cycloheximide analogues cause preinfection resistance to Puccinia graminis var. tritici in spring wheat. Phytopathology 47: 14.
3. HAMILTON, J. M., M. SZKOLNIK, and E. SONDHEIMER. 1956. Systemic control of cherry leaf spot fungus by foliar sprays of Acti-dione derivatives. Science 123:1175.
4. KORNFELD, E. C., R. G. JONES, and T. V. PARK. 1949. The structure and chemistry of Acti-dione, an antibiotic from Streptomyces griseus. Jour. Amer. Chem. Soc. 71: 150.
5. PHELPS, W. R., J. E. KUNTZ, AND A. J. RIKER. 1957. Antibiotics delay oak wilt symptoms on inoculated northern pin oaks in Central Wisconsin. Phytopathology 47: 27.
6. WHIFFEN, A. J. 1948. The production, assay, and antibiotic activity of Acti-dione, an antibiotic from Streptomyces griseus. Jour. Bacteriology 56: 283.

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EFFORTS TO CONTROL CROWN GALL OF ROSES WITH ANTIBIOTICS

Peter A. Ark and W. S. Sibray

Crown gall of roses (caused by Agrobacterium tumefaciens) is a serious disease in many rose nurseries in California and is especially prevalent in establishments where rotation is either limited or non-existent. Since the crown gall organism is strictly a wound parasite, the practices that lead to injury to the rootstock are of considerable importance. Many nurserymen provide for callus formation on rose cuttings after removing the buds from the part of the stem that will be underground. The cuttings are then inserted in well-prepared soil to the depth of four or more inches. The soft callus tissue is easily bruised and scratched when a callused cutting is pushed through the mulch paper and then rubbed against the sandy or rocky constituents of the soil. The obvious injuries produced in this manner provide very convenient portals of entry for crown gall bacteria.

To prevent the occurrence of the disease in roses in the nursery, a program of antibiotic dip treatments was carried out in 1955 and 1956 in a rose nursery where the annual loss from crown gall had been large. Commercial cuttings of two varieties, Paul's Scarlet and Manetti, non pre-callused, were used in 1955 (Table 1), and, in addition, those of Dr. Huey and Burr Multiflora in 1956 (Table 2). The streptomycin solutions used in 1955 contained 100 and 200 ppm, and the tetracycline solutions, 50 and 100 ppm. In the 1956 trials, the steptomycin dips contained 250, 300, 500, and 600 ppm, respectively, and streptomycin dusts, formulated in Nuclay pyrophyllite, contained 1000 and 2000 ppm. A penetrant composed of 1% glycerine and 1% dipotassium phosphate was used in combination with the streptomycin solutions. To promote better callusing of the cuttings, indole acetic acid (100 ppm) was added at the time of dipping the cuttings. The treatments were performed in a closed, unheated barn, and the duration of each treatment was 2 hours.

Table 1. Experimental control of crown gall on two varieties of roses in Alameda County, California, summer 1955. Plants were dug and read on August 19, 1955.

Treatment	Number of plants dug		Number of plants with crown gall		Percent disease		Percent disease compared with check		Percent control	
	Manetti	Paul's Scarlet	Manetti	Paul's Scarlet	Manetti	Paul's Scarlet	Manetti	Paul's Scarlet	Manetti	Paul's Scarlet
No treatment (check)	179	23	99	22	55.3	95.7	100	100	0	0
Streptomycin dip										
100 ppm	283	12	37	5	13.1	41.7	23.7	43.5	76.3	56.5
200 ppm	332	44	15	28	4.5	63.6	8.1	66.4	91.9	33.6
Tetracycline dip										
50 ppm	314	18	116	11	36.9	61.1	66.7	63.8	33.3	36.2
100 ppm	317	30	58	17	18.2	56.7	32.9	59.2	67.1	40.8

In the Burr Multiflora rootstock, very little crown gall developed under conditions which resulted in a considerable amount of the disease in the Manetti and Paul's Scarlet varieties. Thus, out of 558 Burr Multiflora plants dug in the check, there were only 6 plants with small galls, a 1.07 percent incidence of the disease. In the plot with the penetrant and another plot with 600 ppm streptomycin, plus penetrant, there was only one diseased plant in each of the 2 lots of 554 and 560 plants examined. The Burr Multiflora appears very resistant to crown gall under field conditions and should be considered for use where there is no objection to this stock.

Streptomycin dips up to 200 ppm gave some control of crown gall (Table 1). With Manetti rose, control was obtained with streptomycin and tetracycline dips, while with Paul's Scarlet commercial control was not so good.

There is significant difference in susceptibility of rose cuttings to crown gall, Paul's Scarlet being the most susceptible (Table 2).

Table 2. Experimental control of crown gall of roses in Alameda County, California, summer 1956. Number of rose cuttings planted for each series of the test was 600 with the exception of the Paul's Scarlet variety, for which the numbers varied from 300 to 600. Plants dug and examined August 15, 1956.

Treatment	Number of plants dug			Number of plants with crown gall			Percent disease			Percent disease compared with check			Percent control		
	Manetti	Paul's Scarlet	Dr. Huey	Manetti	Paul's Scarlet	Dr. Huey	Manetti	Paul's Scarlet	Dr. Huey	Manetti	Paul's Scarlet	Dr. Huey	Manetti	Paul's Scarlet	Dr. Huey
No treatment — Check 1	462	164	535	144	106	105	31.1	64.6	19.6	100.0	100.0	100.0	0	0	0
Streptomycin dip															
250 ppm	522	130	544	18	39	38	3.4	30.0	6.9	10.9	46.3	35.2	89.1	53.7	64.8
300 ppm	510	94	541	64	65	24	12.5	69.1	4.4	40.1	107.0	22.4	59.9	0	77.6
500 ppm	385	128	498	13	24	37	3.4	18.7	7.4	10.9	28.9	37.7	89.1	71.1	62.3
600 ppm	545	117	416	52	22	60	9.7	10.2	14.4	31.1	15.8	73.4	68.9	74.2	26.6
250 ppm plus P*	528	240	548	107	123	49	20.3	51.2	9.1	89.0	87.9	71.6	11.0	12.1	28.4
300 ppm plus P	380	165	554	33	44	15	8.7	26.6	2.7	38.0	45.7	21.2	62.0	54.3	78.8
500 ppm plus P	467	108	560	96	58	6	20.5	53.7	1.08	89.9	92.2	8.5	10.1	7.8	91.5
600 ppm plus P	534	146	539	26	30	18	4.9	20.5	3.3	21.5	35.2	25.9	78.5	64.8	74.1
*Penetrant only (P) — Check 2	549	67	552	125	39	70	22.8	58.2	12.7	100.0	100.0	100.0	0	0	0
Streptomycin-Nuclay dust															
1000 ppm	539	85	554	117	55	86	21.7	64.7	15.5	69.8	100.0	79.0	30.2	0	21.0
2000 ppm	575	104	522	137	28	87	23.8	26.9	16.7	76.5	41.8	85.2	23.5	58.2	14.8

It appears likely that different rose varieties absorb the streptomycin differently. The penetrant used in the experiments seems to retard the movement of the antibiotic in rose cuttings, in spite of the fact that its constituents have been reported to favor penetration and movement of streptomycin in plants[1, 2]. The streptomycin dusts (Table 2) containing the active streptomycin up to 2000 ppm did not give significant control of crown gall on the rose cuttings. Although there was considerable variation in the action of the streptomycin in the different tests, it appears that significant control of crown gall was obtained in Manetti when a dip of 500 ppm streptomycin for 2 hours was used.

It should be mentioned that both streptomycin and tetracycline antibiotics moved into all parts of the cuttings and produced varying degrees of chlorosis on the first few leaves of the opening buds, but this did not show up in the subsequent foliage and the plants did not seem to show any other signs of distress.

Summary

Crown gall, caused by Agrobacterium tumefaciens, causes significant losses in rose cuttings planted in soils heavily infested with the crown gall bacteria. The portals of entry for the organism are wounds in the callus tissue resulting from procedures used in the propagation of rootstocks.

Of the varieties used in the experiments, Paul's Scarlet was found to be the most susceptible and Burr Multiflora the most resistant.

While some control of crown gall was obtained in the Manetti and Dr. Huey cuttings with 2-hour streptomycin dips containing 500 and 600 ppm, respectively, no satisfactory control was shown by streptomycin dusts (1000 and 2000 ppm). Tetracycline (100 ppm) dips showed 67 percent control in Manetti and only 40 percent control in Paul's Scarlet.

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[1] Ark, Peter A., and Stanley M. Alcorn. 1956. Antibiotics as bactericides and fungicides against diseases of plants. Plant Dis. Reptr. 40:85-92.

[2] Gray, R. A. 1955. Increasing the effectiveness of streptomycin against the common blight of bean with glycerin. Plant Dis. Reptr. 39: 567-568.

CONTROL OF DOWNY MILDEW OF CUCUMBER WITH ANTIBIOTICS

Peter A. Ark and James B. Thompson[1, 2]

Several reports have appeared in the literature on the capacity of streptomycin for controlling downy mildew of cucumber caused by Pseudoperonospora cubensis, downy mildew of tobacco caused by Peronospora tabacina, and downy mildew of lima bean caused by Phytophthora phaseoli. A brief report on the effect of other antibiotics on downy mildew of cucumber has been made by the authors. Since some antibiotics may be of practical importance by virtue of nontoxicity to plants and of better performance than that of many conventional sprays, it was thought that experimental data on a considerable number of less well-known antibiotics would be of interest.

METHODS

For the tests on effects on spore germination, sporangia were washed off the surface of infected leaves and kept in a saturated atmosphere overnight with water and with various concentrations of the antibiotics, after which they were placed in sterilized Syracuse glasses and observed for the discharge of the swarmspores. Observations were made at 2, 6, 18, and 24-hour intervals, and the combined results were used to compute percentages of germination (Table 1).

For inducing the disease on the experimental plants, sporangia were obtained in the same way as for the germination tests. The plants were sprayed with water suspension of the sporangia and held in a humid moist chamber over night, with subsequent incubation under open-bench greenhouse conditions for an average of 7 days before the records of the disease were taken.

To measure the effect of the antibiotics on the disease, counts were made of the lesions on a total of 24 cucumber leaves on 6 plants for each treatment in the trials, and from this the average number of lesions per leaf was determined (Tables 2 and 3). The plants were sprayed with a water suspension of sporangia, then after 15 minutes, with the antibiotic (Table 2), or first with the antibiotic, then 60 minutes later with the inoculum (Table 3).

RESULTS

Complete inhibition of germination of sporangia of Pseudoperonospora cubensis was obtained when aureomycin 500 ppm, filipin 100 ppm, gramicidin S 500 ppm, patulin 100 ppm, and tyrothricin 100 and 500 ppm, were applied (Table 1). Nearly complete inhibition resulted when endomycin 500 ppm, mycostatin 500 ppm, polymixin 100 ppm, and terramycin 100 ppm, were used. Albamycin 100 ppm, bacitracin 500 ppm, and chloromycetin 500 ppm, did not cause any appreciable lowering of germination, while gramicidin (Dubos), 500 ppm, neomycin 500 ppm, and streptomycin 100 ppm, permitted 36, 36, and 31 percent germination, respectively, with 89 percent germination in the check with water only.

When the antibiotics were sprayed on plants either prior to (Table 3) or following (Table 2) the inoculation with the swarmspores, satisfactory control of downy mildew of cucumber was obtained with amphomycin (500 ppm), mycostatin (500 ppm), patulin (100 ppm), polymixin (100 ppm), streptomycin (1000 ppm) Nuclay dust, terramycin (500 ppm), and tyrothricin (100, 300, and 500 ppm). Comparisons between the two methods of application, before or after inoculation, can be made only for a few of the antibiotics and there appeared to be no significant difference.

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[1] Professor of Plant Pathology and Senior Laboratory Technician, respectively.

[2] The authors express their gratitude to the following firms: Merck, Sharp and Dohme for streptomycin, griseofulvin, gramicidin S, tyrothricin, and bacitracin; Chas. Pfizer and Co. for terramycin; Pennick and Co. for amphomycin, albamycin, endomycin, and neomycin; Olin Mathieson Co. for mycostatin; Lederle Co. for tetracycline; Glaxo Co. of England for griseofulvin; and Upjohn Co. for filipin.

Table 1. Effect of antibiotics on germination of sporangia of Pseudoperonospora cubensis.

Antibiotic, ppm	Percent germination after 24 hours	
	Untreated check	Treated
Aureomycin, 500	89	0
Albamycin, 100	89	84
Bacitracin, 500	69	62
Chloromycetin, 500	89	78
Endomycin, 500	89	3
Filipin, 100	68	0
Gramicidin (Dubos), 500	89	36
Gramicidin S, 500	89	0
Griseofulvin, 880, plus 1% dimethyl formamide*	42	57
Griseofulvin, 440, plus 1% dimethyl formamide*	42	40
Mycostatin, 500	89	1
Neomycin, 500	89	36
Patulin, 100	89	0
Polymixin, 100	89	2
Streptomycin, 100	89	31
Terramycin, 100	89	1
Tetracycline, 500	89	1
Tyrothricin, 100	89	0
*Dimethyl formamide (check)	42	51

Table 2. Control of downy mildew (Pseudoperonospora cubensis) of cucumber with antibiotics. The plants were sprayed with the inoculum and 15 minutes later with the antibiotic.

Antibiotic, ppm	Number of lesions per leaf		Percent control
	Untreated check	Treated	
Endomycin, 500	16.2	2.10	87.10
Gramicidin (Dubos), 500	4.7	2.60	44.70
Gramicidin S, 500	4.7	0.11	97.00
Griseofulvin, 440	43.6	33.13	23.60
Mycostatin, 500	43.6	4.70	89.20
Patulin, 100	43.6	0.00	100.00
Polymixin, 100	16.2	5.50	66.10
Terramycin, 100	43.6	13.18	68.30
Terramycin, 200	43.6	14.00	67.90
Terramycin, 500	43.6	6.30	85.60
Tetracycline, 100	16.2	14.70	9.30
Tetracycline, 200	16.2	10.50	35.20

Table 3. Control of downy mildew (Pseudoperonospora cubensis) of cucumber with antibiotics. The plants were sprayed with the antibiotic, followed by the inoculum in about 60 minutes.

Antibiotic, ppm	Number of lesions per leaf		Percent control
	Untreated check	Treated	
Amphomycin, 500	14.6	0.08	99.46
Amphomycin, 300	14.6	0.50	96.58
Albamycin, 100	26.5	11.50	56.60
Bacitracin, 500	4.7	1.70	63.80
Chloromycetin, 200	26.8	14.40	43.30
Endomycin, 500	68.2	0.38	99.45
Filipin, 100	4.7	0.80	83.00
Filipin, 50	4.7	1.50	68.10
Filipin, 25	4.7	2.50	46.80
Gramicidin (Dubos), 500	68.2	64.20	5.90
Gramicidin S, 500	68.2	0.33	99.52
Gramicidin S, 400	34.1	5.20	85.10
Gramicidin S, 200	34.1	7.50	78.10
Griseofulvin, 440	26.8	16.20	39.60
Mycostatin, 500	26.4	3.10	88.30
Patulin, 100	68.2	0.28	99.59
Polymixin, 100	56.3	7.30	87.10
Streptomycin dust, 1000 (in Nuclay)	68.2	8.30	87.80
Terramycin, 500	68.2	3.20	95.30
Terramycin, 200	26.8	7.00	73.90
Terramycin, 100	26.8	8.20	69.40
Tetracycline, 100	56.3	17.20	69.50
Tetracycline, 200	26.5	5.40	79.50
Tyrothricin, 500	14.6	0.08	99.46
Tyrothricin, 300	14.6	0.58	96.03
Tyrothricin, 100	14.6	1.58	89.18

Literature Cited

1. ARK, PETER A., and JAMES P. THOMPSON. 1956. Action of certain antibiotics on downy mildew of cucumber. (Abstr.). Phytopathology 46: 634.
2. COE, DONALD M. 1955. Streptomycin as a control for downy mildew of cucurbits. Plant Dis. Reptr. 39: 729-730.
3. GROSSO, JOHN J. 1954. Control of tobacco blue mold by antibiotics. Plant Dis. Reptr. 38: 333.
4. ZAUMEYER, W. J., and R. E. WESTER. 1956. Control of downy mildew of lima beans with streptomycin. (Abstr.). Phytopathology 46: 32.

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CONTROL OF DOWNY MILDEW OF LETTUCE IN THE EVERGLADES[1]

R. S. Cox

Abstract

Excellent control of lettuce downy mildew (Bremia lactucae) was obtained over a two-year period with zineb and during one year with maneb. Other materials were either less effective or were objectionable because of phytotoxicity. The importance of coverage from the standpoint of disease control, and the role phytotoxicity plays in respect to yield and quality are discussed.

The Everglades area of south Florida appears to be well suited to lettuce production from several standpoints. The rich, muck soils with their controlled water tables constitute a desirable substrate for the plant. Also, the low rainfall and cool weather usually prevailing during the 5-month period from December through April provide further environmental conditions conducive to lettuce growth and head formation. In addition, proximity of the Everglades to the densely populated Northeastern States should insure a stable market for the produce.

Nevertheless, the lettuce industry plays a relatively minor role in the agricultural economy of the Everglades area. Surveys showed that this is due largely to two factors: (a) difficulty in producing a crop, and (b) poor keeping quality of the produce in transit, hence a poor reputation at market terminals. These surveys further revealed that several diseases figure prominently both in production and in keeping qualities of the produce in transit. One of the most important of these diseases is downy mildew, caused by Bremia lactucae (1).

During periods of cool, wet weather, downy mildew can be devastating in its action, resulting in complete abandonment of extensive acreages. Under less severe conditions, quality of the produce is impaired through destruction of the lower (wrapper) leaves and subsequent reduction of head size. Efforts on the part of the grower to control the disease have not been satisfactory. This is somewhat surprising since fungicidal control of this disease has been reported from other areas on lettuce grown under glass or in open seed beds (5, 6, 7). No record of control was found, however, on field-grown lettuce. During the past two years an effective program has been developed for control of downy mildew in the Everglades area (1, 2). This report is a summary of that work.

MATERIALS AND METHODS

Two tests were run: one in 1955 at Shawnee Farms, Moore Haven; the other in 1956 at the Everglades Experiment Station. The Great Lakes No. 118 variety was used in both tests. Fifty-foot single-row plots were employed in a 7 x 7 latin square design in 1955 and in randomized blocks (four replications) in 1956. In 1955, 12 sprays were applied, approximating a semi-weekly schedule from January 11 through February 15. For the 1956 test, 15 sprays were applied at approximately 5-day intervals during the period January 27 to March 29.

Applications were made with the experimental spray rig described elsewhere (3) employing a pressure of 300 p. s. i. Two Myer's jumbo nozzles with #3 orifices were used directly over the row until head initials were formed. After that, the arrangement consisted of one nozzle directly overhead, and another dropped to either side of the plant to within about 3 to 6 inches of the ground level and placed in a position parallel to the soil line. Based on 18-inch rows, gallonage output per acre ranged up to 150.

Disease severity, phytotoxicity and plant vigor were scored on a 0-11 basis where 11 represented maximum rating. Yield data were obtained only in 1955. The heads were graded according to size and designated as 3's, 4's, and 5's according to commercial standards. (The number represents the dozens of heads in that particular grade necessary to fill a standard crate.) Yield was converted to monetary returns per acre based on the formula,

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[1]Florida Agricultural Experiment Station Journal Series, No. 614.
The author gratefully acknowledges the assistance of Nelson Nelms, Field Foreman.

$$\sum YK\left(\frac{G-C}{N}\right) - S$$

where:

Y = number of heads of a grade per treatment
G = current market price of grade per crate (market price on February 1, 1955 for No. 3's, $4.50; No. 4's, $5.00; No. 5's, $3.50)
N = number of heads per crate for each grade
C = packing cost per crate
K = conversion factor to acreage
S = total cost of spray operation per acre.

EXPERIMENTAL RESULTS

1955 Test: -- Downy mildew was present in epiphytotic proportions at the time of the first application and continued so throughout the course of the test. Results appear in Table 1 and Figures 1 and 2. Several fungicides showed significant control. Zineb (Dithane Z-78) was outstanding. This is in agreement with results obtained in seed bed trials in this country (5) and with lettuce grown under glass in England (6, 7). Tribasic copper sulfate and chloranil (Spergon) gave effective degrees of control, but were phytotoxic. Copper toxicity has been reported before on lettuce when applied as a soil drench in North Carolina (4), and as a foliage spray in England (6, 7). PCNB was ineffective fungicidally and was severely phytotoxic. Significant correlations were not obtained when either disease severity or phytotoxicity alone was compared with vigor or yield. As one might expect, however, a high correlation (93 percent) was obtained between the combined effects of disease and phytotoxicity, expressed as vigor (Fig. 1), and yield (Fig. 2). Chemical injury as well as disease appeared to reduce head size. Where phytotoxicity occurred in the absence of disease control (PCNB plots), the heads on the average were smaller than in other plots. Where phytotoxicity was severe and disease control intermediate, the head size was correspondingly intermediate (tribasic copper sulfate and chloranil). The heads were still larger in plots where disease control was intermediate in the absence of phytotoxicity (captan and ferbam), and largest where disease control was best in the absence of phytotoxicity (zineb).

1956 Test: -- Downy mildew was much less severe in 1956 than in 1955. Zineb and the other "bis" carbamates, maneb (Manzate) and nabam plus zinc sulfate, provided superior control. Captan and streptomycin sulfate (Agristrep) in mixture gave a significant degree of control. This is interesting in view of the fact that neither of these materials was effective when used alone. Lack of control with streptomycin is of further interest since this material has been shown to be effective against other downy mildews, particularly blue mold of tobacco. Ferbam and tribasic copper sulfate gave a significant degree of control, but appeared to be less effective than in 1955 when they were used at a higher concentration (compare Tables 1 and 2). Chloranil was ineffective at the concentration used.

PCNB, chloranil, and tribasic copper sulfate again caused injury even though the concentration was reduced over that of the previous year (Tables 1, 2). Streptomycin also caused injury in the form of marked chlorosis and stunting, and nabam plus zinc sulfate caused a contact burn and slight stunting.

DISCUSSION

Based on this work, it would appear that the growers' failure to control downy mildew in past years was due in all likelihood to inadequate spray operations. Most growers were using nozzles only at the overhead position throughout the season. Inferior disease control would be expected in such cases because of poor coverage of the lower leaf surface. Also, their gallonage output was on the order of 100 or less per acre. This generally is inadequate with closely spaced (18-inch rows) leafy vegetables under south Florida conditions.

This work also points out the important role phytotoxicity plays in the evaluation of the relative effectiveness of fungicides. Both tribasic copper sulfate and chloranil provided appreciable control in 1955, but the calculated monetary return from plots treated with either of these materials was actually less than that of the check (Table 1). This obviously was due

Table 1. Comparative effect of various fungicides on development of lettuce downy mildew, host plant response, and yield. Shawnee Farms, Moore Haven, Florida, 1955.

Treatment[a]	Disease severity	Phyto-toxicity	Vigor	Yield (number) 3's	4's	5's	Total	Monetary return per acre
Check	7.6	0.0	6.0	1.4	15.1	10.1	26.6	$868.00
Dithane Z-78	2.1**	0.0	9.3**	8.4	22.7	5.0	36.1**	1354.00
Orthocide 50-W	3.4**	0.0	7.7*	3.3	23.4	6.7	33.4*	1174.00**
Fermate	3.3**	0.0	8.0**	3.4	20.3	10.4	34.1*	1137.00*
Tribasic Copper Sulfate	3.1**	4.5**	5.4	1.1	14.0	12.0	21.1	786.00
Spergon	3.9**	5.5**	4.9	0.7	12.6	12.6	25.9	758.00
PCNB	7.6	5.0**	3.1**	0.0	3.3	8.0	11.3	262.00
L.S.D. .05	0.5	1.3	1.3	---	---	---	6.0	209.00
.01	0.7	1.8	1.8	---	---	---	8.1	278.00

[a] All materials were used at concentrations of 3 pounds / 100 gallons except for Spergon and Tribasic Copper Sulfate; these were used at 4pounds/100 gallons. Active ingredients of the materials as follows: Dithane Z-78, 65% zineb; Orthocide 50-W, 50% captan; Fermate, 76%ferbam; Tribasic Copper Sulfate, 53% metallic copper; Spergon, 48% chloranil; PCNB, 75% pentachloronitrobenzene.
* Significant at the 5% level. ** Significant at the 1% level.

Table 2. Comparative effect of various materials on development of lettuce downy mildew and on host plant response. Everglades Experiment Station, Belle Glade, Florida, 1956[a].

Treatment	Concentration ppm or pounds/100 gallons	Severity of downy mildew Mar. 20, 1956	Phytotoxicity Mar. 20, 1956	Vigor Mar. 20, 1956
Check	---	3.2	0.0	7.0
Manzate	1 1/2 pounds	0.2**	0.0	9.0
Parzate	2 pounds	0.5**	0.0	10.2
Parzate plus Agristrep	2 pounds plus 100 ppm	0.2**	2.0[b]	9.9
Dithane D-14 plus Zinc sulfate	2 quarts plus 3/4 pounds	0.8**	3.0[c]	8.5
Manzate plus Agristrep	1 1/2 pounds plus 100 ppm	1.0**	1.0[d]	9.2
Orthocide plus Agristrep	2 pounds plus 100 ppm	1.5**	0.5[d]	9.9
Fermate	2 pounds	2.0*	0.0	10.2
Tribasic Copper Sulfate	2 pounds	2.2*	3.0[e]	8.7
Tribasic Copper Sulfate plus Agristrep	2 pounds plus 100 ppm	2.2*	1.0[d]	9.8
Fermate plus Agristrep	2 pounds plus 100 ppm	2.2*	2.5[d]	9.0
Spergon	2 pounds	2.8	2.0[e]	9.2
Agristrep	100 ppm	3.0	2.0[b]	8.5
Orthocide	2 pounds	3.0	0.0	8.2
PCNB plus Agristrep	2 pounds plus 100 ppm	3.2	3.0[b]	8.0
Agristrep	200 ppm	3.5	3.5[b]	8.5
PCNB	2 pounds	3.8	3.0[e]	8.2
L.S.D. .05		1.0	0.9	
.01		1.4	1.2	

[a] Active ingredients of materials not listed in Table 1: Manzate, 70% maneb; Parzate, 65% zineb; Agristrep, 37% streptomycin sulfate; Dithane D-14, 19% nabam.
[b] Injury in the form of chlorosis and stunting of plant.
[c] Small necrotic spots and stunting of plant. [d] Chlorosis but not stunting. [e] Stunting but no chlorosis.
* Significant at the 5% level. ** Significant at the 1% level.

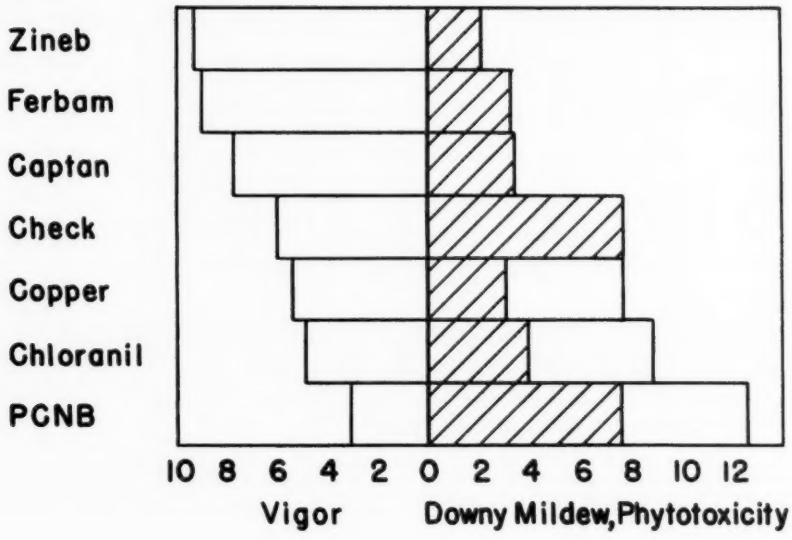

FIGURE 1. Cumulative effect of downy mildew and phytotoxicity on vigor of lettuce plants. The open portion of the bar to the right of the vertical line represents phytotoxicity; the hatched portion, downy mildew.

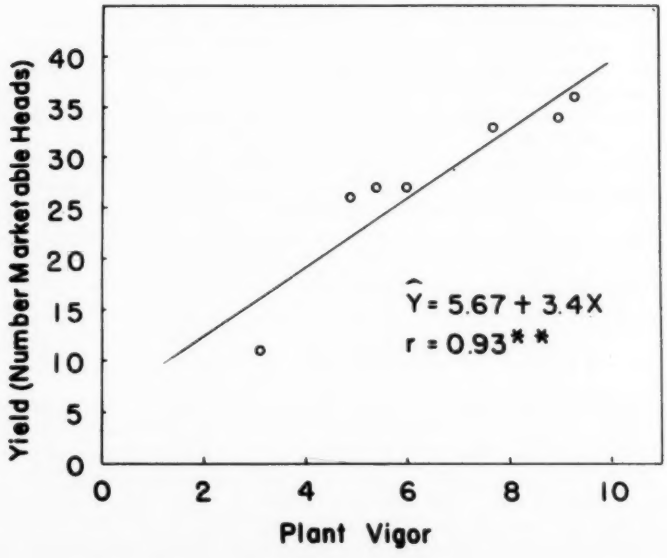

FIGURE 2. Regression of yield of marketable lettuce heads on plant vigor. Plant vigor is the composite effect of downy mildew and phytotoxicity (see Fig. 1).

to the adverse effect of phytotoxicity on yield. It is sobering to realize that at least one large grower had been spraying with chloranil for mildew control for several years. In 1956, nabam plus zinc sulfate, generally considered safe on leafy vegetables, caused a contact burn on the lower leaves and appeared to stunt the plants. Heads from these plots were consistently smaller than those from plots sprayed with zineb or maneb. With the present emphasis on quality of produce, closer scrutiny of the more subtle effects of chemical injury on crop plants would appear to be in order.

Literature Cited

1. COX, R. W. 1955. A preliminary report on diseases of lettuce in the Everglades and their control. Plant Dis. Reptr. 39: 421-423.
2. COX, R. S. 1956. Control of downy mildew of lettuce. (Abst.). Phytopath. 46: 10.
3. COX, R. S., D. S. HARRISON, and C. S. YAGER. 1955. A versatile spray rig for small field plots. Plant Dis. Reptr. 39: 48-50.
4. ELLIS, D. E., and R. S. COX. 1951. The etiology and control of lettuce damping off. North Carolina Agr. Exp. Sta. Tech. Bul. 94.
5. HAASIS, FRANK A., and D. E. ELLIS. 1950. Effect of fungicidal drenches on incidence of lettuce downy mildew in the seed bed. Plant Dis. Reptr. 34: 310-311.
6. POWLESLAND, RUTH, and W. BROWN. 1954. The fungicidal control of lettuce downy mildew caused by Bremia lactucae. Ann. Appl. Biol. 41: 461-469.
7. WILD, H. 1947. Downy mildew disease of the cultivated lettuce. Trans. Brit. Mycol. Soc. 31: 112-125.

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PREVENTION OF DECAY OF STRAWBERRIES WITH POST-HARVEST TREATMENTS[1, 2]

G. R. DiMarco and B. H. Davis

Summary

Several chemicals were tested in 1955 to determine their value in controlling Botrytis and Rhizopus rots of harvested strawberries. Captan at 1200 ppm (active) and Mycostatin (Nystatin), an antifungal antibiotic, at 100 ppm (active) were selected for use in a hydrocooler in 1956. A vacuum-cooled treatment was also included. A substantial reduction in decay was obtained with Mycostatin added to the cooling water. Captan was less effective in controlling decay than Mycostatin and it left an objectionable residue. Vacuum-cooled berries showed a high percentage of mold in these tests, probably due to slight injury of the berries caused by the treatment.

Screening tests were conducted in 1955 with a number of chemicals, used as dips, to determine their value in controlling Botrytis and Rhizopus rots of harvested strawberries. The most effective materials were added to the cooling water (32° F) in a hydrocooler in 1956. The great amount of interest among farmers and farmers' cooperatives in hydrocooling fruits and vegetables prompted this method of applying the chemicals. Hydrocooling of strawberries on an experimental basis in 1955 resulted in an increase in rots. The 1956 tests were conducted with the hope that the chemicals would control the rots and that the advantages of removing the field heat by hydrocooling could be retained. Vacuum cooling, which is also receiving considerable interest, was also used. The results of these tests are reported.

MATERIALS AND METHODS

In the 1955 tests quart baskets of strawberries of the Sparkle variety were dipped in solutions and suspensions of the chemicals at room temperature, drained, and stored at room temperature and at 42° F. The chemicals and concentrations used in these screening tests are given in Table 1. No marked reduction in rots or marked differences between treatments were obtained and the specific results are not reported. Captan (1200 ppm active) and Mycostatin[3] (100 ppm active), an antifungal antibiotic, were considered the most promising in these tests. These materials were used in the cooling water in a 25 cubic-foot pilot model hydrocooler in 1956. The berries were cooled for 20 minutes. A pilot model vacuum cooler (chamber evacuated by a steam-operated, 3-stage, air ejector) was also used. The berries were cooled for 15 minutes at 29 inches of vacuum. Dry berries and berries hydrocooled without the additon of chemicals served as checks.

Four tests were conducted on field-run strawberries during the 1956 strawberry season. The berries obtained were considered typical of those which can be expected to reach the market during a given season. The first lot of strawberries was relatively green and one lot was slightly overripe. Each lot of berries received was progressively poorer in quality; the last lot being the poorest with considerable insect damage present as well as some decay.

Two 16-quart containers were used per treatment in each test. Fiber board containers were used for the vacuum-cooled treatment and wire-bound crates for the hydrocooled treatments. One of these was stored at room temperature under a tarpaulin and the other at 42° F.

The average temperature of the berries before and after the 20-minute treatment in the hydrocooler was 74° and 36° F, respectively. The average temperature under the tarpaulin was 79° F and the average humidity 85 percent. Within the refrigerator the average temper-

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[1] Paper of the Journal Series, New Jersey Agricultural Experiment Station, Rutgers University, the State University of New Jersey, Department of Plant Pathology, New Brunswick.

[2] This work was conducted with the cooperation of the departments of Food Technology and Agricultural Economics.

[3] Supplied by E. R. Squibb and Sons.

Table 1. Chemicals tested in 1955 for control of decay of strawberries.

Chemical	Concentration of active chemical ppm
Aureomycin	200, 400
Captan	1200, 2400, 3600
Dowicide A-M245	400, 1000
Keepsit*	20% solution
Mycostatin	100
Peracetic acid	1000
Phytomycin	1000
Pyridinethione (Na salt)	500
Sodium Propionate*	10% solution
Sorbic acid	2500, 5000
Streptomycin sulfate	1000
Trichloromelamine	400

* Concentration in percent

ature and humidity was 42° and 100 percent respectively. After vacuum cooling for 15 minutes, a thermometer inserted to the center of the berries gave an average reading of 35°. This, however, was not the true temperature of the unpunctured berries. Since the bulb of the thermometer within the berry becomes moistened with the fruit juices, and since cooling under vacuum is by evaporation, the recorded temperature was much lower than the actual temperature of the sound berry.

One crate of berries was inspected and evaluated at the beginning of each test to determine the percentage of good berries at the outset. Berries held at room temperature were inspected daily. Those held under refrigeration were inspected every other day. Berries were graded into three grades: 1) berries that met USDA inspection requirements; 2) berries which did not meet these requirements but showed no visible mold; and 3) berries with visible surface mold. Data were taken on 2 quarts from the top and 2 quarts from the bottom layer of each crate. Data were taken on a weight basis. The graded berries were discarded after each inspection.

RESULTS

The data given in Figures 1, 2, 3, and 4 are averages of the results of all four tests. The average percentage of good berries (those meeting USDA inspection requirements) before treatment was 79. The results given express the percentage of the original good berries remaining in good condition at the time data were taken. The data showing the amount of decay with no visible mold present are not represented in the figures. The data on the top and on the bottom layers showed that the berries in the bottom layer decayed more rapidly than those in the top layer. The data from the two layers were combined to give the results reported.

Hydrocooling alone resulted in an increase in decay compared with the dry check. Presumably the residual moisture on the berries after hydrocooling provided a favorable condition for growth and spread of the fungi present.

The refrigerated untreated berries kept well. Since only a few test crates were placed in an efficient refrigerator these berries could be expected to keep better than those stored commercially where large quantities would be cooled less rapidly.

Although captan gave some degree of control, it left a visible residue on the berries, especially those at the bottom of the quart containers. This would be very objectionable from the marketing viewpoint.

Vacuum cooling gave poor control of decay. A high incidence of visible mold resulted at both room and refrigerated temperatures. The vacuum cooling is believed to have injured surface cells which would favor the development of decay organisms.

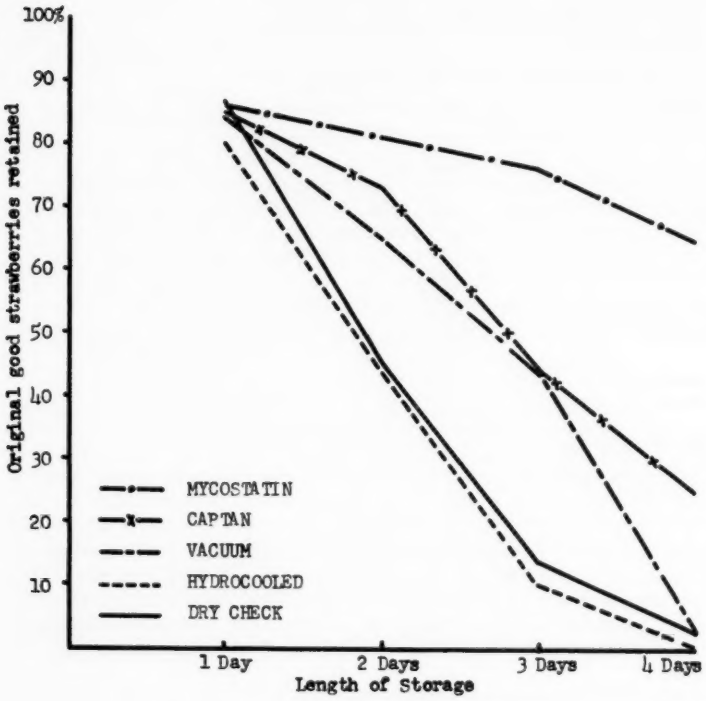

FIGURE 1.
Average retention of original good strawberries in four tests after various post-harvest treatments and storage at room temperature.

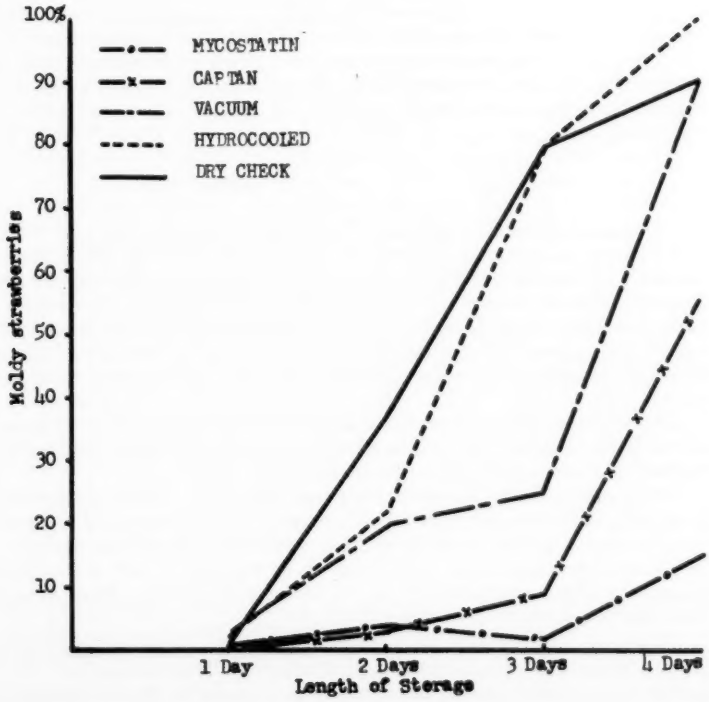

FIGURE 2.
Average percent of moldy strawberries in four tests after various post-harvest treatments and storage at room temperature.

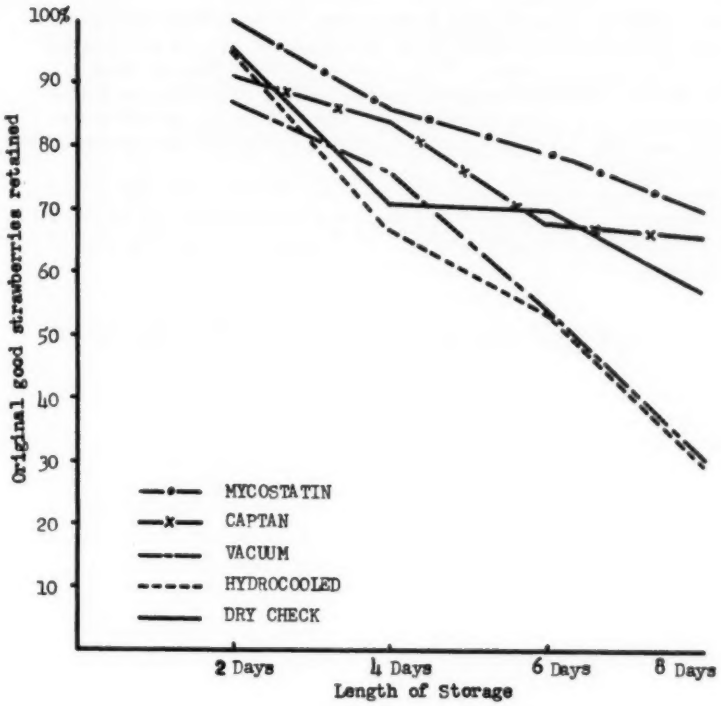

FIGURE 3.
Average retention of original good strawberries in four tests after various post-harvest treatments and storage at 42° F.

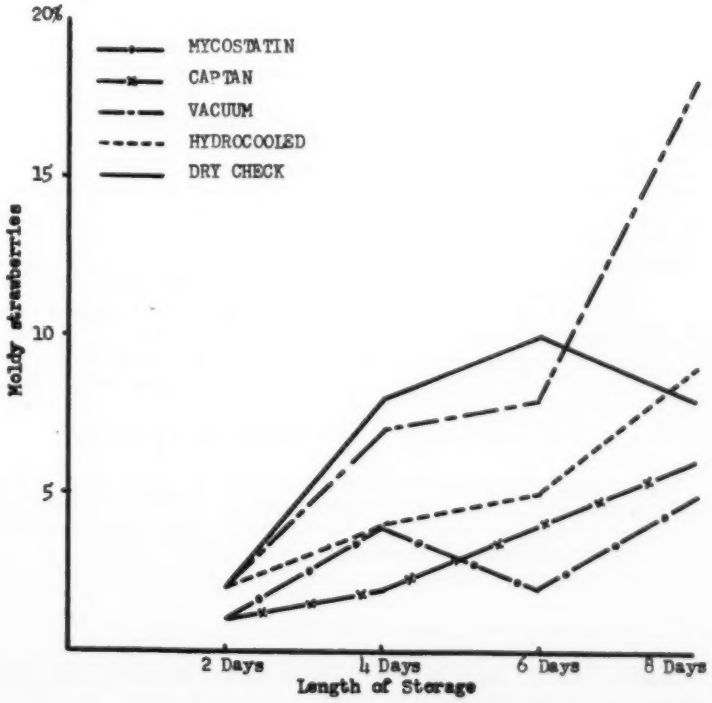

FIGURE 4.
Average percent of moldy strawberries in four tests after various post-harvest treatments and storage at 42° F.

The best control in these tests was obtained with Mycostatin. After 8 days storage at 42° F, Mycostatin-treated berries graded 70 percent good as compared with 30 percent and 57 percent good for the hydrocooled and dry check, respectively. After 4 days at room temperature 65 percent of the Mycostatin-treated berries were healthy as compared with 0 and 3 percent for the hydrocooled and dry check respectively. Also, at room temperature after 4 days there were 100 percent and 90 percent molded berries in the two checks compared with 15 percent molded in the treated berries.

Mycostatin has also shown promise in extensive tests conducted for two seasons on the control of post-harvest peach decays[4]. Its practical application as a post-harvest treatment for peaches and strawberries has not yet been determined. Such questions as cost, stability, formulations, and residue tolerances have not yet been completely answered.

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[4] DiMarco, G. R. and B. H. Davis. Prevention of decay of peaches with post-harvest treatments. Plant Dis. Reptr. 41: 284-288. 1957.

COTTONSEED OIL FORMULATIONS OF ORGANIC FUNGICIDES FOR TOBACCO

Saul Rich and Gordon S. Taylor

Summary

Mixtures of cottonseed oil with wettable powder suspensions of six organic fungicides were applied to shade tobacco foliage with a mist blower. The oil formulations left very little visible residue in the field. Maneb, zineb, dichlone, and streptomycin gave no visible leaf damage, Thioneb caused moderate leaf damage, while the thiram paste formulation severely damaged sprayed leaves. The incidence of blue mold was not severe enough to test adequately the protective capacity of the spray treatments. The disease data indicated, however, that the cottonseed oil may not reduce the protection against blue mold (Peronospora tabacina) afforded by zineb, dichlone, and thiram. The oil may reduce the protective action of maneb, streptomycin, and Thioneb.

After the leaves were processed, they were examined for spray residue, and tested for burn quality. Only the zineb-oil treated leaves showed any residue after processing. The thiram and Thioneb treated leaves were of poor burn quality. The streptomycin-oil treated leaves gave off a strong bitter aroma when burned. The other treatments had little effect on burn quality.

Cottonseed oil may be safely mixed with certain organic fungicides for spraying tobacco foliage. It is possible that these formulations may be used successfully on other crops.

INTRODUCTION

In recent years, there have been outbreaks of blue mold (Peronospora tabacina) on tobacco in the field. Growers have used fungicidal dusts because dusts are easier to apply than sprays and can be formulated to leave little residue. Most plant diseases in the Northeast, however, are better controlled by sprays. This probably results from the better sticking ability of dried spray residues during the periods of rainfall when the fungicide is most needed. But because sprays stick so well, they may leave visible residues which could show on the processed leaf. This is particularly undesirable for wrapper tobacco. In addition, water suspensions are not well suited for application from mist blowers and aircraft which must be used when the tobacco becomes too tall for the use of ground equipment. The smaller droplets tend to evaporate before reaching the plants, leaving the fungicide to drift about in a condition unsuited for sticking to the leaf surface. This latter disadvantage has been overcome in the application of insecticides by mixing with petroleum oils. Organic fungicides, however, are usually not compatible with the ordinary spray oils.

It was decided, therefore, to test mixtures of organic fungicides with cottonseed oil. The choice of cottonseed oil was based on the work of Clayton, et al. (1). These workers found that cottonseed oil was safe to use on tobacco with copper oxide. In addition, they reported that the oil itself made tobacco more resistant to blue mold, even though cottonseed oil was not fungitoxic to the blue mold organism.

MATERIALS AND METHODS

All materials were applied with an improved model of the wheelbarrow mist blower first described by Potts, et al. (2), using 3 times the standard concentrations of formulations. The rates applied, in pounds of formulation per 100 gallons, were maneb, zineb and Thioneb at 6, thiram paste at 9, and dichlone at 3 pounds. Streptomycin was applied at 600 ppm. Three quarts of cottonseed oil and 2 ounces of Triton B-1956 per 100 gallons were added to each fungicide suspension.

The experimental area contained 10 rows of Connecticut 49 tobacco, each row 33 feet long, growing in a shade tent. Each plot, which was duplicated, consisted of 5 rows of 4 plants each. The treatments were randomized in each of the two replicates. Sprays were applied on July 20 and July 26 when the plants were 3 to 4 feet tall and actively growing. The

leaves were thoroughly wetted with spray. Counts of total blue mold lesions in each plot made on July 31 and August 6. The sprayed leaves were picked on September 1. The picked leaves were cured at the Tobacco Laboratory with other shade tobacco. Fermentation was done in a commercial bulk. After being processed, the leaves were examined for visual spray residue, and tested for burn quality. In the burn tests, 4 burns were made on each of 10 leaves at random from each batch. These burn tests were done by M. R. Goldsmith.

RESULTS

The cottonseed oil formulation left much less residue on the leaves in the field than normally occurs with standard hydraulic applications of the same materials. The thiram mixture severely injured the leaves, while the Thioneb mixture caused moderate leaf injury. None of the other treatments caused visible injury in the field.

Blue mold lesions recorded on July 31 were undoubtedly the result of infections which took place before the first sprays were applied. Estimates of protection could only be had by following the progress of the disease as shown by differences between the two counting dates. Only the second replicate was near enough to the infection source to show a difference between counts.

Table 1. Results of cottonseed oil plus organic fungicide treatments on tobacco.

Fungicide	Leaf injury	Visible residue on cured leaves	Burn quality
Maneb	0	0	Good
Zineb	0	Slight	Fair
Dichlone	0	0	Good
Streptomycin	0	0	Harsh odor
Thiram	Severe	0	Poor
Thioneb	Moderate	0	Poor
Check	0	0	Good

The disease advance was not sufficient to give an adquate test of the protection against blue mold afforded by these mixtures. There was an indication, however, that the cottonseed oil may not reduce the effectiveness of zineb, dichlone and thiram against blue mold. It also appeared that cottonseed oil may reduce the fungitoxicity of maneb, streptomycin and Thioneb.

After being processed, only those leaves which had been sprayed with the zineb mixture showed any visible residue, and that was very slight. The results of the burn test are given in Table 1. It is interesting to note that thiram and Thioneb, the two treatments which caused injury in the field, resulted in tobacco with very poor burn quality. The streptomycin mixture appeared to reduce the holding quality of the ash and made for a strong, bitter aroma. The zineb treatment reduced the holding quality of the ash. The maneb and dichlone treatments did not influence burn quality appreciably.

CONCLUSION

These tests demonstrate that cottonseed oil may be safely mixed with certain organic fungicides for spraying tobacco foliage. It is possible that these formulations may be used on other crops where oil formulations of organic fungicides are desired.

Literature Cited

1. CLAYTON, E. E., J. E. SMITH, K. J. SHAW, J. G. GAINES, T. W. GRAHAM, AND C. C. YEAGER. 1943. Fungicidal tests on blue mold (Peronospora tabacina) on tobacco. Jour. Agric. Res. 66: 261-276.
2. POTTS, S. F., and R. A. SPENCER. 1947. A small portable mist blower for applying concentrated spray. U. S. Bur. Ent. and Pl. Quar. ET-234: 1-3.

DEPARTMENT OF PLANT PATHOLOGY AND BOTANY
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TREATMENT OF VEGETABLE SEED FOR IMPROVED EMERGENCE -- 1956[1]

V. R. Wallen[2], J. K. Richardson[3], L. Cinq-Mars[3] and W. Bell[4]

Identical vegetable seed treatment trials for improved emergence and the prevention of damping-off and seed rot were conducted at three separate locations: in Ontario at Ottawa and St. Catharines, and in Quebec at St. Jean. In the trials nine treatment materials, alone or in combination, and 13 different kinds of vegetables were used. In germination tests on filter paper 80 percent or more of the seed of each vegetable germinated. Insecticides, fungicides, and combination insecticide-fungicide treatments were used.

The treatment materials and their chemical composition are as follows:

Aldrin 50W -- 50% technical aldrin
Arasan -- 50% thiram
Arasan 75 and Aldrin 50W (2:1) -- 50% thiram + 16.7% technical aldrin
Delsan AD -- 60% thiram + 12.75% technical dieldrin
Orthocide 75 -- 75% captan
Ortho Seed Guard -- 50% captan + 16.5% gamma isomer of benzene hexachloride
Panogen 15 -- 2.2% cyano (methylmercuri) guanidine
Phygon -- 50% dichlone
Spergon -- 97% chloranil
Spergon and Aldrin 50W (3:1) -- 72.75% chloranil + 12.5% technical aldrin
Thioneb 50W -- 50% polyethylene thiuram sulphides

As in other trials (1) the seed was treated in 2 ounce lots for the small-seeded vegetables and up to 1 pound samples for the larger seeded kinds at the rates indicated in Table 1. The treated seeds were stored at room temperature in stoppered flasks at 65° to 75° F for 2 to 3 weeks before being sown in the field. The seed of each treated lot was sown in replicate rod row plots in randomized blocks. An identical scheme of randomizing the plots within the 13 blocks was followed at each location. Emergence counts were made 2 weeks after sowing or as soon as possible after this time. Tables 2, 3, and 4 show the percent emergence of the various kinds of seeds after treatment with the various materials at the three locations.

It is apparent from Tables 2, 3, and 4 that all kinds of seeds responded to seed treatment by increased emergence, with the exception of swede seed which showed no statistically significant increase at the three locations. It is also apparent that there was no significant increase with corn seed at St. Catharines. Bush beans and lima beans treated with Ortho Seed Guard showed greater emergence than did seed given other treatments at Ottawa and St. Catharines, while Orthocide 75 was the best treatment at St. Jean. For soybeans, Ortho Seed Guard, Orthocide 75, and Arasan 75 plus Aldrin 50W were the best treatments at St. Catharines, St. Jean, and Ottawa. Higher emergences were obtained when beet seed was treated with Phygon, Arasan, and Panogen than when it was given any other treatment, both at Ottawa and St. Jean. The cucurbits cucumber, muskmelon, pumpkin, squash, vegetable marrow and watermelon responded to a number of seed treatments, only a few of which gave no significant increases. For cucumber, watermelon, and vegetable marrow at all three locations significant increase in emergence occurred with all fungicide and fungicide-insecticide combinations. Orthocide 75 was the best treatment for peas at Ottawa and St. Jean while Ortho Seed Guard was the best treatment at St. Catharines. Climatic conditions influenced emergence considerably; the colder soil in the spring at St. Jean and Ottawa apparently was unfavorable for germination as most control plots had considerably lower emergence than similar plots at St. Catharines.

The addition of an insecticide greatly benefited emergence and health of plants at Ottawa and St. Catharines in all bush bean treatments. Although in most cases the addition of an insecticide did not improve the emergence of bean seed; the insecticide in combination with a

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[1]Contribution No. 1603 from the Botany and Plant Pathology Division, Science Service, Canada Department of Agriculture, Ottawa, Ontario.
[2]Plant Pathologist[3], Associate Plant Pathologist[4], Technician.

Table 1. Dosages of seed treatment materials per 100 lb. of seed for various kinds of seed used in trial.

Kind of seed	Aldrin 50W	Arasan	Arasan 75 and Aldrin 50W (2:1)	Delsan	Orthocide 75	Ortho Seed Guard	Panogen 15	Phygon	Spergon SL	Spergon SL and Aldrin 50W (3:1)	Thioneb 50W
Bush Bean	1 oz.	2 oz.	3 oz.	3 oz.	2-1/2 oz.	3 oz.	4 oz.	2 oz.	3 oz.	4 oz.	4 oz.
Beet	1 oz.	8 oz.	12 oz.	8 oz.	12 oz.	12 oz.	4 oz.	8 oz.	16 oz.	4 oz.	16 oz.
Corn	1 oz.	5-1/3 oz.	3 oz.	4-1/2 oz.	1-1/2 oz.	3 oz.	4 oz.	1-1/2 oz.	3 oz.	4 oz.	3 oz.
Cucumber	1 oz.	3 oz.	3 oz.	3 oz.	3 oz.	3 oz.	4 oz.	4 oz.	6 oz.	4 oz.	8 oz.
Lima Bean	1 oz.	3 oz.	3 oz.	3 oz.	2-1/2 oz.	3 oz.	4 oz.	2 oz.	3 oz.	4 oz.	4 oz.
Muskmelon	1 oz.	3 oz.	3 oz.	3 oz.	3 oz.	3 oz.	4 oz.	3 oz.	6 oz.	4 oz.	6 oz.
Pea	1 oz.	3 oz.	3 oz.	3 oz.	2-1/2 oz.	3 oz.	4 oz.	2 oz.	3 oz.	4 oz.	4 oz.
Pumpkin	1 oz.	3 oz.	3 oz.	3 oz.	2 oz.	3 oz.	4 oz.	4 oz.	6 oz.	4 oz.	8 oz.
Soybean	1 oz.	2 oz.	3 oz.	3 oz.	3 oz.	3 oz.	4 oz.	2 oz.	3 oz.	4 oz.	4 oz.
Squash	1 oz.	3 oz.	3 oz.	3 oz.	2 oz.	3 oz.	4 oz.	4 oz.	6 oz.	4 oz.	8 oz.
Swede	1 oz.	4 oz.	3 oz.	4 oz.	1-1/2 oz.	3 oz.	4 oz.	4 oz.	8 oz.	4 oz.	8 oz.
Veg.Marrow	1 oz.	3 oz.	3 oz.	3 oz.	2 oz.	3 oz.	4 oz.	4 oz.	6 oz.	4 oz.	8 oz.
Watermelon	1 oz.	3 oz.	3 oz.	3 oz.	2 oz.	3 oz.	4 oz.	4 oz.	6 oz.	4 oz.	8 oz.

Table 2. Mean percentage field emergence from vegetable seed treated with various seed treatments, based on four replicates of 100 seeds each. (Ottawa)

Kind of seed	Untreated	Aldrin 50W	Arasan	Arasan 75 and Aldrin 50W (2:1)	Delsan AD	Orthocide 75	Ortho Seed Guard	Panogen 15	Phygon	Spergon SL	Spergon SL and Aldrin 50W (3:1)	Thioneb 50W	L.S.D.[a]
Bush bean	24	13	63.5[c]	65.25	69.25	82.75	85.0	67.5	75.75	66.75	48.5	71.5	5.82
Beet	36.75	26.75	128.25	73.75	76.75	98.5	71.5	105.5	106.5	40.25	22.25	99.25	23.2
Corn	76.25	77.0	91.25	92.75	89.75	93.75	91.5	93.0	86.0	85.75	87.75	83.25	3.28
Cucumber	5.25	8.75	39.25	55.25	61.25	55.25	62.5	49.5	44.75	43.25	56.75	47.0	5.71
Lima Bean	16.0	12.5	52.25	61.75	60.25	59.0	70.25	59.5	45.5	20.25	30.5	27.75	6.31
Muskmelon	30.25	52.25	41.75	74.75	79.25	51.75	61.25	35.5	43.0	40.0	60.25	43.5	7.09
Pea	20	23.5	74.0	70.75	74.5	83.75	76.0	68.25	77.75	63.25	50.5	70.5	5.84
Soybean	32.0	31.75	61.25	72.0	61.0	70.75	67.25	64.25	54.0	47.5	52.25	55.5	5.09
Squash	61.75	68.25	63.0	77.0	72.25	78.75	85.75	79.0	83.0	54.25	65.75	78.5	6.95
Swede	71.75	68.75	82.25	74.0	71.25	79.0	74.75	77.25	78.25	73.5	76.25	77.25	----[b]
Veg.Marrow	17.25	15.0	68.0	69.25	58.75	72.75	62.0	66.75	73.0	43.0	35.25	73.25	5.19
Watermelon	12.75	17.25	50.75	75.0	70.25	58.5	69.5	50.25	53.75	51.25	58.25	52.5	5.88
Pumpkin	79	82.25	87.0	84.25	87.0	86.25	88.75	86.75	86.25	79.0	82.75	90.5	3.88

[a] L.S.D. at the 5% level

[b] No significant increase between treatments

[c] Underlined numbers indicate significant increase in emergence at 5% level.

Table 3. Mean percentage field emergence from vegetable seed treated with various seed treatments, based on four replicates of 100 seeds each. (St. Catharines)

Kind of seed	Untreated	Aldrin 50W	Arasan	Arasan 75 and Aldrin 50W (2:1)	Delsan AD	Orthocide 75	Ortho Seed Guard	Panogen 15	Phygon	Spergon SL	Spergon SL and Aldrin 50W (3:1)	Thioneb 50W	L.S.D.[a]
Bush Bean	63.25	63.5	65.25	79.25[c]	74.5	59.25	84.0	65.5	61.5	76.75	79.25	74.5	10.13
Beet	78.0	86.5	83.0	77.5	93.25	91.25	87.0	78.5	79.5	54.0	39.5	48.25	----[b]
Corn	88.5	85.25	83.75	84.5	87.75	81.25	74.75	75.25	85.75	86.5	85.5	94.75	----[b]
Cucumber	21.25	25.0	34.25	33.5	47.5	36.75	45.0	48.5	34.0	42.25	54.5	53.25	9.83
Lima Bean	39.0	38.5	60.0	60.0	68.5	66.75	86.75	59.25	69.75	79.0	65.5	76.75	11.08
Muskmelon	23.25	25.5	33.25	47.25	61.0	36.75	54.75	61.5	51.5	59.5	70.5	61.75	8.63
Pea	56.0	61.0	52.0	69.75	74.0	56.0	81.25	61.0	56.25	58.75	73.75	65.25	11.69
Pumpkin	68.25	83.5	68.0	83.0	83.25	73.25	95.75	86.5	76.0	81.0	84.5	84.0	6.17
Soybean	61.25	52.75	76.25	77.75	73.75	68.25	80.0	64.25	68.5	69.0	76.0	69.5	6.23
Squash	47.25	69.5	51.5	62.75	69.5	62.5	83.5	73.0	79.25	74.25	85.25	82.25	12.23
Swede	62.0	62.0	54.75	40.25	44.5	56.5	60.0	63.5	60.0	55.5	63.0	67.25	----[b]
Vegetable Marrow	47.75	54.75	57.5	54.0	64.75	58.75	68.75	61.0	62.0	64.75	62.75	62.25	6.46
Watermelon	24.0	43.25	41.25	57.75	54.5	48.0	76.0	63.75	59.5	65.75	62.75	58.5	11.5

[a] L.S.D. at the 5% level

[b] No significant increase between treatments

[c] Underlined numbers indicate significant increase in emergence at 5% level.

Table 4. Mean percentage field emergence from vegetable seed treated with various seed treatments, based on four replicates of 100 seeds each. (St. Jean)

Kind of seed	Untreated	Aldrin 50W	Arasan	Arasan 75 and Aldrin 50W (2:1)	Delsan AD	Orthocide 75	Ortho Seed Guard	Panogen 15	Phygon	Spergon SL	Spergon SL and Aldrin 50W (3:1)	Thioneb 50W	L.S.D.[a]
Bush bean	36.25	8.25	72.0[c]	61.5	65.25	84.5	76.25	62.75	77.0	70.5	56.75	73.5	7.64
Beet	39.5	41.0	67.25	46.25	41.0	60.25	39.5	89.5	74.0	17.75	13.25	59.5	14.82
Corn	78.0	80.5	85.0	90.75	79.25	88.5	84.25	84.25	86	84	78.5	87.5	----[b]
Cucumber	23.0	25.5	48.25	53.0	47.0	61.25	43.5	64.0	51.75	55.75	38.75	48.25	9.25
Lima Bean	40.5	12.25	47.75	53.0	42.25	63.5	59.5	54.5	60.75	44.5	34.5	42.0	7.9
Muskmelon	19.0	18.5	52.0	53.0	65.25	56.5	58.75	51.5	48.0	40.0	44.25	45.5	7.98
Pea	45.25	38.5	77.0	65.5	78.25	88.0	81.5	66.25	82.0	72.5	59.75	78.25	9.57
Pumpkin	71.0	66.25	67.5	82.5	74.0	80.75	79.25	80.0	83.0	75.5	69.25	81.75	4.78
Soybean	37.5	32.75	58.75	65.25	54.75	66.0	60.75	48.25	57.0	49.0	54.5	49.25	11.58
Squash	58.0	55.0	51.75	51.25	50.0	84.25	81.0	79.0	84.0	45.75	23.25	80.5	9.46
Swede	60.25	65.0	68.5	68.0	72.0	65.5	76.25	73.25	73.0	70.25	72.25	70.5	----[b]
Vegetable Marrow	23.75	22.5	60.75	53.25	50.0	61.0	56.0	61.5	64.5	46.0	45.0	56.5	6.14
Watermelon	6.5	9.0	62.5	46.25	33.0	66.25	67.75	70.0	63.0	53.75	43.75	69.5	8.5

[a] L.S.D. at the 5% level

[b] No significant increase between treatments

[c] Underlined numbers indicate significant increase in emergence at 5% level.

fungicide produced a high emergence together with excellent control of seed corn maggot. In both the control and fungicide plots, infestation by the seed corn maggot resulted in 9.25 to 25.0 percent of baldhead plants while the highest infestation with any treatment that included an insecticide was 2.75 percent. Aldrin did not improve emergence but effectively controlled the seed corn maggot.

It is obvious from this work that the use of an insecticide incorporated with a fungicide is beneficial in the control of seed corn maggot in all bean crops and is recommended where this insect is a problem. Although bush beans, soybeans, and lima beans were the only crops attacked by this insect in this test the use of a combination insecticide-fungicide treatment as a precaution against seed corn maggot is recommended for corn, peas, and cucurbit crops. The use of an insecticide alone is not recommended as in almost every case (Tables 2, 3 and 4) no increase in emergence was noted.

Literature Cited

1. WALLEN, V. R., and A. J. SKOLKO. 1953. Treatment of vegetable seed of low germination. Plant Dis. Reptr. 37: 66-68.

BOTANY AND PLANT PATHOLOGY LABORATORY, SCIENCE SERVICE, CANADA DEPARTMENT OF AGRICULTURE, OTTAWA, ONTARIO

EFFECTIVENESS OF COPPER-TREATED PAPER WRAPPERS IN PREVENTING SPREAD OF RHIZOPUS NIGRICANS IN MATURE-GREEN TOMATOES

Edward E. Butler

Summary

In three tests copper-treated fruit wraps (Hartman wraps) failed to reduce the spread of Rhizopus nigricans from fruit to fruit in green-wrap tomatoes significantly. In in vitro tests Rhizoctonia solani, Oospora lactis f. parasitica, Fusarium sp., Alternaria tenuis, Botrytis cinerea, and Pleospora sp. grew almost unimpaired through the copper-treated paper, whereas the copper greatly restricted the penetration of Phytophthora capsici and P. drechsleri.

Tomatoes harvested in Central California during September and October supply a number of eastern markets. Fruits are picked mature-green ("green-wraps") (8), wrapped individually in a thin paper, and packed tightly in wooden lug boxes. A smaller volume of fruit is packed unwrapped in wire-bound crates.

A considerable portion of the green-wrap fruit is shipped by rail to points as distant as New York City and thus tomatoes may be en route as long as 10 to 15 days (4). During this period fruit may be subject to rot caused by a number of fungi (3). Potentially, the most destructive species are those which grow through the paper wrappers and infect adjacent healthy fruit. A number of fungi have this capacity (1, 3, 5): Botrytis cinerea, Sclerotium rolfsii, Phytophthora spp., and Rhizopus nigricans.

Recently there has been an interest in the possibility that the incidence of rot incurred through the action of spreading fungi could be reduced or eliminated by wrapping tomato fruit in chemically treated paper. A search of the literature revealed only two reports on the control of decay in tomatoes by the use of chemically treated wraps. Both of these were by R. G. Tompkins in England. He pointed out that a wrap effective in inhibiting fungus growth must contain a slowly volatile inhibitor. Tompkins obtained good results in reducing decay by wrapping green tomatoes in paper treated with a mixture of iodine and potassium iodide (6). Iodine wraps did not damage tomato fruit. In later studies (7) he obtained a marked reduction of mold growth on stored tomatoes through the use of wraps impregnated with O-phenyl-phenol plus hexamine.

Neither of the foregoing fruit wraps are available commercially in the United States. Of immediate interest to California shippers and growers is the Hartman fruit wrap currently used extensively in the Western States for the prevention of spread of Botrytis cinerea in pear storage. The Hartman wrap contains basic copper carbonate with minor amounts of basic copper sulfate and copper hydroxide. The total amount of copper expressed as metallic copper is 1.3 percent[1].

This report presents the results of tests to determine the suitability of the Hartman wrap for prevention of fungus spread in California green wrap tomatoes.

STUDIES WITH FRUIT

Rhizopus nigricans was selected as the test organism. It not only grows rapidly from fruit to fruit under optimum conditions but infected fruits become watery and their contents "leak". Lugs of such fruit arriving at markets are recognized as "wets" and are generally degraded in value. A chemically treated paper wrap designed to prevent fungus spread would necessarily have to restrict the movement of R. nigricans.

In the first test, four lugs of mature green tomatoes, variety Pearson, were packed October 6, 1956, in a commercial packing house at Tracy, California. Fruits in two lugs were wrapped in the Hartman wrap and fruit in two others in untreated paper. Each lug contained three layers of approximately 35 fruits each in rows 7 long x 5 wide. Spores of Rhizopus

[1] The writer wishes to express his appreciation to the Crown Zellerbach Corp. for supplying the Hartman wraps and reporting their chemical composition, and to the Potlatch Forests, Inc. of Pomona, California, for furnishing untreated fruit wraps.

Table 1. Amount of fruit rot caused by Rhizopus nigricans in tomato fruits wrapped in copper-treated as compared with untreated paper wraps[a].

Kind of wrapper Test No.	Total number of fruits in lugs	Percent of fruit with rot: In individual lugs	Average for test
Copper treated:			
Test No. 1[b]	102	6.8	
	94	4.3	
			5.6
Test No. 2[c]	110	27.2	
	92	23.9	
	56	33.9	
	60	35.0	
			28.9
Untreated:			
Test No. 1[b]	104	6.7	
	100	11.0	
			8.8
Test No. 2[c]	110	31.8	
	92	28.3	
	56	35.7	
	70	21.4	
			29.3

[a]Fruit packed in lugs; the four corner fruits in the center layer of each lug inoculated with a spore suspension of R. nigricans.
[b]Test No. 1 — 9 days at 73° F.
[c]Test No. 2 — 10 days at 55°, 3 days at 68°, and 3 days at 79° F, total 16 days.

Table 2. Growth of several tomato-rot fungi into sterile agar blocks separated by copper-treated paper from similar blocks inoculated with the respective fungi, as compared with controls separated by untreated paper.

Fungus tested	Mycelial growth in test block (as percent of control)
Rhizoctonia solani Kuehn	100
Oospora lactis (Fres.) Sacc. f. parasitica Pritchard & Porte	100
Fusarium sp.	90
Rhizopus nigricans Ehr.	90
Alternaria tenuis auct.	80
Botrytis cinerea Pers.	80
Pleospora sp.	70
Phytophthora capsici Leonian	10
Phytophthora drechsleri Tucker	0

nigricans from an aqueous suspension of 14 isolates were inoculated into the four corner fruits in the second layer. The results are summarized in Table 1.

R. nigricans failed to spread extensively from the four infection centers in each lug to all of the surrounding fruit. Of the 24 fruits in each lug in direct contact with the inoculated tomatoes less than half were infected. Nevertheless the copper-impregnated wrap failed to prevent spread completely. Fruits did not ripen uniformly and a few of those in direct contact with Rhizopus-rotted fruit were green. This may account in part for the lack of infection and spread since Rhizopus sp. has been reported to attack only ripe injured fruit. However, in the present tests most of the fruits were ripe. Actually very little is known concerning the factors affecting spread of Rhizopus nigricans or other pathogens from fruit to fruit in tomatoes.

Test number 2 was initiated October 19, 1956, with mature green tomatoes harvested at Davis, California. Fruits were divided into four lots in which half were wrapped in the Hartman wrap and half in an untreated commercial paper. They were then packed in appropriate size boxes and inoculated as in the first test. An attempt was made here to simulate temperatures which might occur during transit. Thus fruit were stored at 55° F for 10 days, at 68° for 3 days, and then removed to 79° for 3 more days -- a total of 16 days.

The results of these tests are summarized in Table 1. R. nigricans did not spread rapidly at 55° F. Only a few of the fruits immediately in contact with the inoculated fruits were rotted at 10 days. At this time rots caused by other organisms were not significantly present. During the 3 days at 68°, R. nigricans spread only to a few other fruits near the infection centers. However, at 79° F the fungus spread extremely fast so that one-third to one-half of the fruits were attacked by R. nigricans. The copper-impregnated paper offered no barrier to the spread of R. nigricans.

Another test was made with fruit picked on November 3, 1956, at Davis. By this date some of the fruits were infected in the field, principally by Botrytis cinerea, Alternaria tenuis, and Stemphylium spp. Fruit were immersed one minute in dimethyl di-dodecenyl ammonium chloride, washed in water, dried and wrapped in copper-treated and untreated paper. The test consisted of four treatments, each containing 70 fruits and replicated seven times making a total of 490 fruits for each treatment. Treatments and the average percentage of rotted fruit after 10 days at 70° F were as follows: Wrapped in copper paper and inoculated as in test No. 1 with R. nigricans, 70.4; wrapped in untreated paper and inoculated, 76.3; wrapped in copper paper and uninoculated, 44.8; wrapped in untreated paper, uninoculated, 60.6.

The increase in rot between the inoculated and uninoculated treatments was due to the spread of R. nigricans. This spread was not significantly retarded by the Hartman wrap.

PENETRATION OF COPPER-TREATED PAPER IN VITRO BY TOMATO-ROT FUNGI

At the conclusion of tests with fruit it seemed desirable to know if Rhizopus nigricans and other fungi known to cause tomato rot could penetrate the copper-treated paper of the Hartman wrap. Qualitative in vitro tests on this point were made as follows.

The organisms listed in Table 2 were grown in Petri dishes on potato dextrose agar (PDA) 0.5 cm thick, until mycelium covered the plate. Blocks of agar 2.0 cm square containing mycelium were removed and placed singly in a sterile Petri plate. A 6.0-cm square of copper-treated paper was then centered over the agar block containing the mycelium, and a sterile block of PDA 2.0 cm thick was placed on top of the paper. Controls consisted of untreated paper and tests were made in triplicate. The amount of mycelium in the top agar block was recorded as an estimated percent of the control after 72 hours at 25° C.

As shown in Table 2 most of the fungi grew nearly unimpeded through the copper paper. The two species of Phytophthora were an exception; both are apparently sensitive to copper. Phytophthora drechsleri failed to make any noticeable growth on the blocks over copper paper while a dense mycelium completely covered the control blocks. These observations suggest that the Hartman wrap might effectively prevent the spread of Phytophthora spp. from fruit to fruit.

The results of other tests by the writer substantiate the results reported here, namely, that the Hartman fruit wrap cannot be depended upon to reduce the spread of Rhizopus nigricans significantly. A number of tomato pathogens can grow readily through the Hartman wrap (Table 2), and it is likely that the copper content of this wrap is not sufficient to prevent other fungi from spreading.

Literature Cited

1. BAKER, KENNETH F. 1946. An epiphytotic of Rhizopus soft rot of green-wrap tomatoes in California. Plant Dis. Reptr. 30: 20-26.
2. RAMSEY, G. B., and ALICE ALLEN BAILEY. 1929. The development of soil rot of tomatoes during transit and marketing. Phytopathology 19: 383-390.
3. RAMSEY, GLEN B., JAMES S. WIANT, and LACY P. McCOLLOCH. 1952. Market diseases of tomatoes, peppers and eggplants. U. S. Dept. Agr. Agric. Handbook 28 (June).
4. RICHARDSON, H. B., and JOHN P. UNDERHILL. 1955. California fall tomatoes in New York City. Univ. of California Agric. Ext. Serv. Mimeo. publication, 1956.
5. ROSENBAUM, J. 1918. The origin and spread of tomato fruit rots in transit. Phytopathology 8: 572-581.
6. TOMPKINS, R. G. 1934. Iodized wraps for the prevention of rotting of fruit. Jour. Pomology and Hort. Sci. 12: 311-320.
7. TOMPKINS, R. G. 1938. Treated wraps for the prevention of fungal rotting. Rept. British Food Inves. Board, pp 186-189.
8. WRIGHT, R. C., and E. A. GORMAN, Jr. 1940. The ripening and repacking of mature green tomatoes. U. S. Dept. Agr. Circ. 566.

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PEA DISEASES IN MICHIGAN, 1955 AND 1956[1]

John L. Lockwood, D. J. deZeeuw, A. L. Andersen, and D. J. Hagedorn[2]

INTRODUCTION

Surveys were made in 1955 and 1956 to determine the causes, prevalence, and severity of pea diseases in Michigan. Previously Michigan pea fields had not been surveyed systematically for diseases, but observations in the Fremont and Saginaw areas in 1949 and 1954 had revealed that root rots were causing severe damage to peas in most fields.

Three areas of the State were surveyed, each with a different soil type and each growing peas for a different purpose. The Fremont area, located near Lake Michigan, includes parts of Muskegon and Newaygo Counties. In this area peas are grown on light sandy loam and occasionally on muck, and are processed for baby foods. In the Saginaw area, silt and clay loams predominate, and the crop is processed as frozen peas. The pea crop in the Croswell area in Sanilac County near Lake Huron is grown primarily for canning. Soils in this area vary from sandy to clay loams.

The following pea varieties were grown in Michigan in 1955 and 1956: Alaska, Early Perfection, Freezer 37, Freezonian, Glacier, Lincoln, Miracle, Miragreen, Perfected Freezer, Perfected Wales, Perfection, Pluperfect, Thomas Laxton, Victory Freezer, Wando, and Wyola. In nearly all fields peas were grown as part of a 3- or a 4-year rotation with cereals and row crops.

METHODS

Surveys usually were made within a week of harvest. Six to 11 fields were surveyed in each area, with a total of 22 fields visited in 1955 and 25 in 1956. Tops and roots of pea plants in random portions of each field were examined. Both the frequency of diseased plants and severity of symptoms were used in assigning a disease rating to each field visited. The ratings assigned were the consensus of the observers. A disease rating of 0 indicated no disease; 1, trace to slight infection; 2, moderate infection; and 3, severe infection. The average of all the ratings from the individual fields for each disease in each area was used to express disease severity (Table 1). The 1955 survey was made on June 15-16; in 1956 the Fremont area was surveyed on June 28 and the Saginaw and Croswell areas were visited on July 16.

RESULTS

Root rots were by far the most important group of parasitic diseases of peas in Michigan, in both 1955 and 1956. Root rots were found in all but 3 of 22 fields surveyed in 1955. This disease was very severe at Croswell, whereas the crops in most fields in the Fremont and Saginaw areas were judged slightly or moderately diseased. In 1956, root rots were found in each of 25 fields visited; there were marked increases in severity of these diseases in the Fremont and Saginaw areas and severity continued near Croswell. Three of the fields examined in 1956 had not previously been cropped to peas. In 2 of these only slight infection was found, but in the third root rots were very severe.

The root diseases observed in 1955 appeared to be caused by Aphanomyces (Aphanomyces euteiches) and Fusarium (F. solani f. pisi) but neither microscopic examinations nor isolations were made. In 1956, collections of diseased roots were made from 14 of the 25 fields surveyed. Root tissues were examined microscopically for oospores of A. euteiches, but none was found. Tissue isolations were made on agar media and hemp seeds. Cultures of F. solani f. pisi which produced typical symptoms of fusarium root rot in greenhouse tests were isolated from each collection. No cultures of A. euteiches were obtained. From the symptoms in the

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Table 1. Relative severity of pea diseases in 3 areas of Michigan in 1955 and 1956.

Year and disease		Disease severity in indicated area[a]		
		Fremont	Saginaw	Croswell
1955	Root rots	0.9	1.1	2.4
	Virus diseases	1.6	1.4	0.5
	Sclerotinia wilt	0.0	0.4	0.5
	Ascochyta blight	0.3	0.1	0.0
	Bacterial blight	0.2	0.0	0.0
	Downy mildew	0.2	0.0	0.0
	Powdery mildew	0.2	0.0	0.0
	Near wilt	0.2	0.0	0.0
	Frost injury (Non-parasitic)	1.9	1.9	0.8
1956	Root rots	1.9	1.7	2.3
	Virus diseases	0.5	1.3	0.3
	Powdery mildew	0.0	0.4	0.0
	Ascochyta blight	0.4	0.0	0.0

[a] Each value expresses the relative disease severity in each area and was obtained by averaging the individual field disease ratings from 6-11 fields. Thus 0 = no disease; less than 0.5 = trace of infection; 0.5-1.5 = light infection; 1.5-2.0 = moderate infection; greater than 2.0 = severe infection.

field, from microscopic and cultural findings, and from pathogenicity tests of isolates from diseased roots it appeared that F. solani f. pisi was responsible for practically all root rot in Michigan in 1956.

Virus diseases were second in importance to root rots in Michigan pea fields in 1955 and 1956. One-third of the fields visited in 1955 and one-half of those surveyed in 1956 contained virus-infected plants. In both years a very low incidence of virus infection was found at Croswell. Virus diseases were more prevalent in fields near Fremont and Saginaw than at Croswell in 1955, but in 1956 very little virus infection occurred near Fremont. This reduction was believed to be due to spraying for aphid control earlier in that year. The virus diseases appeared to be of several types. Symptom expression indicated that pea streak, pea stunt, mosaic caused by bean virus 2, enation mosaic, and common pea mosaic were present.

Other diseases found in 1955 were Ascochyta blight (Ascochyta spp.), Sclerotinia wilt (Sclerotinia sclerotiorum), bacterial blight (Pseudomonas pisi), downy mildew (Peronospora pisi), powdery mildew (Erysiphe polygoni), and near wilt (Fusarium oxysporum f. pisi r. 2). Except for Ascochyta blight and Sclerotinia wilt each of these diseases was found to a limited extent in 1 field. Ascochyta blight was found in 3 fields and rated slight in each. Sclerotinia wilt was found in 5 fields, being severe in 1 and slight in 4. This disease is considered as potentially important in Michigan.

The year 1956 was remarkable for the near absence of diseases other than root rots and

virus diseases. One of 8 fields surveyed in the Fremont area was severely infected with Ascochyta pisi. Two of 6 fields at Croswell were infected with powdery mildew, 1 severely. No other diseases were found.

Frost injury was prevalent and damaging in 1955, particularly in the Fremont and Saginaw areas. A late frost killed many pea shoots in both early- and late-planted fields and stimulated the production of new shoots. As a result the peas tended to mature at the same time whether the field had been planted early or late. It was not unusual to see fields in which all the plants still had flowers in bloom when pods were ready for harvest.

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A TOMATO PLANT WITH POWDER-PUFF TYPE OF INFLORESCENCES

T. C. Vanterpool

A tomato plant (Lycopersicon esculentum) with deformed inflorescences similar to those described by Young[1] as "broccoli type" on a watermelon plant, was found in a greenhouse near Saskatoon, Saskatchewan, on July 28, 1956. The variety was the greenhouse strain of Sutton's-Best-of-All. It was the first time in over 25 years that the grower had observed such an abnormality in this variety. The single affected plant was staked, about 6 feet tall, had normal leaves, stems and branches, but in place of normal flowers and fruit only pale green, aborted flower rudiments were formed. There was proliferation of these deformed flowerless trusses with a tendency for leaf sprouts to develop in the older trusses (Fig. 1). From a short distance the abnormal growths resembled a small pale green powder puff. On examination under a binocular microscope, the centres of the floral rudiments were found to be composed of enlarged, colorless, shiny, hyperhydric cell masses with slender, hairy, pale green growths, varying in size from about 0.1 mm wide x 1 mm high to 1 mm wide x 10 mm high, towards the outside. It was not possible to assign with certainty a given rudiment to a particular floral part.

FIGURE 1. An old, abnormal truss taken from the original tomato plant, showing a proliferation of sterile floral parts interspersed with leafy shoots.

The abnormal condition was not transferred to healthy tomato plants by mechanical juice inoculations, nor by using aphids as possible vectors. Cuttings from the abnormal plant were rooted and later scions from normal plants were grafted on these abnormal stocks. In four such grafts, normal scions on abnormal stocks continued to produce normal flowers and finally

[1] Young, P. A. 1956. A watermelon plant with broccoli type of inflorescences. Plant Dis. Reptr. 40:1094.

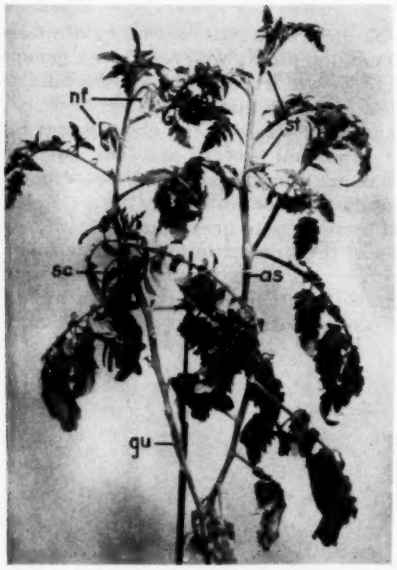

FIGURE 2. An abnormal stock as, with a normal scion sc, 9 weeks after grafting. Graft union, gu; normal flowers, nf; and a sterile truss, st.

FIGURE 3. A closer view of the top part of the plant shown in Figure 2. Normal flowers, nf; and a sterile truss, st.

fruit, while the abnormal stocks continued to produce powder-puff inflorescences (Figs. 2 and 3).

The foregoing results indicate that the teratological condition described on the tomato plant was due to mutation.

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DISEASE INCIDENCE AND YIELDS OF BEANS IN RELATION TO CULTIVATION INJURY IN NORTHEASTERN COLORADO[1]

D. W. Burke and C. E. Seliskar[2]

Abstract

An examination of cultivator damage in 12 bean fields showed that an average of 23.5 percent of the plants had been injured on the lower part of the stem or root. A Fusarium rot of the stele was found to be closely associated with this mechanical damage. Bacterial wilt, caused by Corynebacterium flaccumfaciens, was also favored by this damage. On the other hand, Sclerotinia wilt, caused by Sclerotinia sclerotiorum, was not favored by mechanical damage. Non-injured plants produced a greater seed yield than injured ones in 9 of 10 fields. Application of a herbicide, amine salt of chlorophenoxy butyric acid, killed weeds without showing phytotoxicity to bean plants. However, there was little difference in disease incidence between plots so treated and those receiving mechanical cultivation that had caused relatively little injury.

In many northeastern Colorado bean fields a high percentage of plants are injured on the roots and lower parts of the hypocotyls by mechanical weed control operations. Most of this damage occurs early in the season, before and after plant emergence, when close cultivation methods are used.

It is generally assumed that bean crops sustain little permanent damage from such mechanical injuries. Planting rates have been adjusted to compensate for some reduction in stand. However, it seems reasonable to expect that injured plants are reduced in efficiency even though they survive. Also, it is to be expected that mechanical injuries would be a factor in the incidence of soil-borne diseases. Infection by the bacterial wilt organism, Corynebacterium flaccumfaciens (Hedges) Dowson, was found to be materially increased by injury to the root system (1). Root rots of lima beans, caused by Fusarium solani f. phaseoli (Burk.) Snyder & Hansen, and other pathogens were greater in the presence of mechanical damage by wireworms than in its absence (2). Furthermore, in Colorado in 1955 post-emergence killing of bean plants by the seed corn maggot in some fields appeared to be associated with pre-emergence cultivator injury.

An investigation was made during the 1956 season to determine the extent of cultivator injury in some fields in the Greeley area and its effects on the incidence of soil-borne diseases and on yield. Consideration was given to soil type and cropping history.

METHODS AND RESULTS

Disease Incidence in Injured and Non-Injured Plants

Experimental plots were established in six fields of Pinto beans. Three plots were in light, sandy loam and three in heavy clay loam. One field in each soil type had grown no beans for several years, another was in beans for the second consecutive year, and the third was in beans for the third or more consecutive year.

Some plots in each field were excluded from the usual cultivation operations and were weeded by hand. Two treatments were applied in 4 replications of two 20-foot rows. The first consisted of punching the stems of each plant below soil level to simulate cultivator injury, shortly after plant emergence. In the second, the control, plants were left undisturbed as much as possible.

Only three diseases were fairly common in the fields under observation. They were bacterial wilt, Sclerotinia wilt, caused by Sclerotinia sclerotiorum (Lib.) d By., and a brick-red

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Table 1. Relation of root injury to disease incidence in fields with different cropping histories on two soil types.

Soil type, cropping history, and treatment	Plants affected (percent)[a] with		
	Bacterial wilt	Sclerotinia wilt	Stele rot
Clay loam:			
1st-year beans			
1. Injured	0	0	100
2. Noninjured	0	0	0
2nd-year beans			
1. Injured	0	1	100
2. Noninjured[b]	0	1	9
8th-year beans			
1. Injured	0	90[c]	--[d]
2. Noninjured	0	90[c]	--[d]
Sandy loam:			
1st-year beans			
1. Injured	4.3	0.5	100
2. Noninjured[b]	2.0	3.0	7.5
2nd-year beans			
1. Injured	--[d]	0	100
2. Noninjured[b]	--[d]	0	17.5
3rd-year beans			
1. Injured	5.5	0	100
2. Noninjured[b]	1.0	0	1

[a] Average data from 50 plants in each of 4 replications.
[b] Inadvertent cultivation or hoeing by the farmers caused some mechanical injury.
[c] Estimated infection occurring in the entire plot irrespective of treatment.
[d] Plants were defoliated or killed by Mexican bean beetle or by disease to the extent that data could not be obtained.

to brown discoloration and rot of the stele in roots and stems which yielded principally isolates of Fusarium oxysporum (Schl.) Snyder & Hansen. Typical Fusarium and Rhizoctonia root rots, as evidenced by cortical decay, were very slight in all fields. Damage by seed corn maggots was not observed.

Because of lack of uniformity in stand and maturity within the various plots, yield data were not recorded.

Table 1 shows a comparison of disease incidence in injured and noninjured plants. Fusarium infection of the stele occurred in every plant mechanically injured below the soil line. It was not observed in plants lacking this type of injury. The disease was found to a similar extent in both soil types and appeared to be little affected by cropping history. The height to which stele rot extended beyond the injured area varied from plant to plant. In some plants it reached the first node above the primary leaves.

Bacterial wilt also occurred more frequently on injured plants than on noninjured ones. This disease appeared only in sandy loam.

The occurrence of Sclerotinia wilt, on the other hand, showed no relation to mechanical

Table 2. Incidence of cultivator injury in 12 fields cultivated in various ways.

Field	Plants showing below-ground mechanical injury (percent) in replicate					Cultivation methods
	1	2	3	4	mean	
1	16	16	17	17	16.5	Knives, duckfeet, and hoe [a]
2	31	27	42	36	34.0	Harrow [b], knives and duckfeet, and sinner weeder.
3	37	50	40	34	40.2	Cultro, sinner weeder (in crusted soil), and hoe [a]
4	9	16	7	6	9.5	Cultro, sinner weeder (no crust)
5	15	31	26	17	22.2	Scratcher and rotary hoe
6	18	23	28	10	19.8	Rotary hoe, knives, and duckfeet
7	28	26	35	22	27.8	Rotary hoe, knives, and duckfeet
8	12	9	9	5	8.8	Harrow [b], knives, and duckfeet
9	36	30	13	21	25.0	Rotary hoe, knives, and duckfeet
10	36	17	20	26	24.8	Rotary hoe, knives, and duckfeet
11	40	23	25	38	31.5	Scratcher, rotary hoe, knives, and duckfeet
12	36	12	15	26	22.2	Rotary hoe, knives, and duckfeet

[a] One hoeing by hand.
[b] Harrow was used before the plants emerged.

damage. It appeared mainly in the heavier soil and was most extensive in the field bearing its 8th consecutive crop of beans.

Prevalence of Mechanical Injury in Twelve Fields

Random samples of 100 plants were collected in 4 locations in each of 12 fields at harvest time. The roots were examined for scars of cultivator injuries and type of cultivation methods was recorded for each field. The data obtained are summarized in Table 2.

A large number of plants in all fields received cultivator injury (Figs. 1 and 2). The incidence of such injury was consistently as high where cultivation was close and deep, as when the scratcher and rotary hoe were used. Extent of injury by other methods varied with the farmer, the soil conditions, and possibly other factors. The highest incidence of cultivator injury was found in the ends of the fields. As in past years, bacterial wilt also was most noticeable in the ends of the fields, further indicating the correlation between cultivator damage and bacterial wilt incidence.

In most instances, below-ground mechanical damage and stele rot went hand-in-hand. However, in one field which had borne rye turned under as green manure and a late potato crop in 1955, stele rot was comparatively slight, with 30 percent of the injured plants showing no stele discoloration beyond the point of injury.

Yields of Injured and Noninjured Plants

Seed yields of random samples of 100 injured plants from 10 of the experimental fields were compared with those of 100 noninjured plants (Table 3). In all fields but one, injured

FIGURE 1. Type of cultivator injury occurring commonly in bean fields.

FIGURE 2. Interior of three injured roots showing stele rot as compared with a noninjured root.

Table 3. Yields of plants mechanically injured below the soil line as compared with those of noninjured plants from the same fields.

Field	100 Injured plants	100 Noninjured plants	Difference
	grams	grams	grams
1	1525	2180	655
2	1667	1845	222
3	1690	1965	275
4	2345	2495	150
5	1950	2450	500
6	1223	1493	270
7	1545	1370	-175
8	1290	1970	680
9	1860	2000	140
10	2185	2720	535

plants yielded less than the noninjured ones. It is of interest that in this field (number 7) stele rot was slight as compared with that in other fields. These data would indicate that plants injured by cultivation implements are reduced in yielding ability and that this reduction may result from Fusarium infection of the stele. The importance of bacterial wilt in reduction of yield of injured plants could not be determined because the plants had been cut and windrowed prior to sampling. Their wilted condition generally made recognition of wilt difficult. From the data presented it is difficult to ascertain the practical importance of the lowered yielding ability of injured plants. It remains for further investigation to determine to what extent this is compensated for by more vigorous development of noninjured plants as a result of reduced competition.

Incidence of Root Rot in Chemically Weeded and Cultivated Plots

An experiment was established at Fort Collins to determine in part the effects of chemical weed control on the incidence of root rot in Pinto beans. The purpose was to eliminate root damage occasioned by mechanical cultivation through the substitution of chemical methods for the control of weeds.

Six random replications of four 40-foot rows were sprayed with a herbicide (amine salt of chlorophenoxy butyric acid) at the rate of 1/2 pound per acre, when the beans were in the 2- to 5-leaf stage. Good weed control was obtained and no phytotoxicity to the beans was observed. Except for an occasional hand hoeing late in the season, no mechanical cultivation was applied.

In this test no significant differences in disease incidence were obtained in plots receiving herbicide applications and those cultivated. It should be noted, however, that relatively little cultivator injury was found on the roots of plants in the check plots when compared with that normally found in commercial fields.

Literature Cited

1. FAULKNER, LIN R., and C. E. SELISKAR. 1954. The nature of secondary infection in bacterial wilt of bean caused by Corynebacterium flaccumfaciens (Hedges) Dowson. (Abst.) Colorado-Wyoming Acad. Sci. 4: 50.
2. MIDDLETON, JOHN T., M. W. STONE, and J. B. KENDRICK, Jr. 1949. Incidence of lima bean root rot in soils treated with fumigants and insecticides for control of wireworms. Phytopathology 39: 813-821.

HORTICULTURAL CROPS RESEARCH BRANCH, AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE, AND DEPARTMENT OF BOTANY AND PLANT PATHOLOGY, COLORADO A & M COLLEGE, FORT COLLINS

INCIDENCE OF BACTERIAL PATHOGENS IN DRY BEANS IN IRRIGATED DISTRICTS OF NEBRASKA, WYOMING AND COLORADO IN 1954 AND 1955

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Abstract

Bacterial pathogens were isolated from many discolored seeds concentrated in cull piles from hundreds of acres of dry beans. Normally white seeds of Great Northern varieties were colored differently by each of four pathogens. On this basis it was determined that the yellow bacterial wilt organism, Corynebacterium flaccumfaciens (Hedges) Dowson, and the orange variety, recently named C. flaccumfaciens var. aurantiacum Sch. & Chris., were present in an average of 68 and 26 percent, respectively, of the discolored seeds sampled during 2 years. The common blight bacterium, Xanthomonas phaseoli (E. F. Sm.) Dowson, and the fuscans variety, X. phaseoli var. fuscans (Burk.) Starr & Burk., were found in 5 and 2 percent, respectively. A similar relative distribution from year to year and among locations in Nebraska and Wyoming suggested that the pathogens are spread in wind-blown soil. Yellowed seeds of Pinto beans yielded isolates of all but X. phaseoli var. fuscans. However, the normal pigmentation of these seeds made differentiation of infecting bacteria on the basis of seed discoloration impracticable.

INTRODUCTION

Surveys of the types of relative incidences of bacterial pathogens of beans in the irrigated dry-bean-producing districts near Scottsbluff, Nebraska, Torrington, Wyoming, and Greeley, Colorado, were made in 1954 and 1955. In the hot, dry weather prevailing during most of the growing season, it was often difficult to differentiate plant symptoms produced by one bacterial pathogen from those produced by another. However, as suggested by Schuster and Christiansen (3, 4), cull piles of dry beans at processing establishments provide a source of information on the incidence of bacterial diseases. Seeds from hundreds of acres showing discolorations characteristic of bacterial infections are concentrated in cull piles.

METHODS AND RESULTS

Representative collections of cull beans were obtained from 11 establishments in the 3 districts. Bacterial isolates from many seeds in each collection were examined for cultural characteristics on nutrient agar and potato cylinders as well as for the Gram-stain and morphological characteristics described by Burkholder (1). The principal isolates examined, though not tested for pathogenicity, were found to be typical representatives of four pathogenic types of bacteria.

Among the normally white Great Northern beans the different pathogens were found to be consistently associated with rather specific shades or types of seed discoloration. The bacteria and a standard designation (2) for the approximate seed coloration with which they were associated, respectively, were as follows: the common yellow wilt organism, Corynebacterium flaccumfaciens, lemon yellow to Chinese yellow; the recently reported orange variety, C. flaccumfaciens var. aurantiacum (4), orange peel to carrot red; the common blight organism, Xanthomonas phaseoli, rattan to buff; and the fuscans variety, X. phaseoli var. fuscans, rattan to buff with a darkened area, usually near the hilum.

Distinguishing the pathogens in infected Pinto bean seeds was impracticable because of the dark mottled seed coat.

Another method of separating wilt-infected seeds from those infected by blight organisms was found usable with both Great Northern and Pinto beans. This is based on the fact that seeds infected with the wilt organisms usually yield a much more dense suspension of bacteria

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Table 1. Incidence of 4 bacterial pathogens in samples from collections of infected bean seeds, 1954 and 1955.

Type and source of seeds	C. flaccumfaciens		C. flaccumfaciens var. aurantiacum		X. phaseoli		X. phaseoli var. fuscans	
	1954	1955	1954	1955	1954	1955	1954	1955
Great Northern:[a]								
Minatare, Nebr.	--	66	--	32	--	1	--	1
Scottsbluff, Nebr[b]	69	49	25	45	4	4	2	2
Gerring, Nebr.[b]	75	35	20	55	4	9	1	1
Gerring, Nebr.	43	72	49	28	4	--	4	--
Mitchell, Nebr.	85	70	11	14	2	10	2	6
Morrill, Nebr.[b]	67	72	24	18	6	9	3	1
Torrington, Wyo.	85	88	8	10	3	1	4	1
Average	70.7	64.6	22.8	28.8	3.8	5.7	2.7	2.0
Pinto:[c]								
Torrington, Wyo.	+	--	+	--	+	--	0	--
Greeley, Colo.	+	--	+	--	0	--	0	--
Greeley, Colo.	+	--	+	--	+	--	0	--
Greeley, Colo.	+	--	+	--	+	--	0	--

[a] Incidence expressed as average number (percentage) in 2 to 6 random counts of 100 infected seeds.

[b] Culls from electric-eye sorted beans. All other collections were culls from gravity-and-hand-sorted beans.

[c] Separation on basis of seed coloration was not practicable; + means that the organism was isolated from representative samples and 0 that it was not found.

in water to which cracked seeds are added than do blight-infected seeds. However, this method was little used in the present study.

From each of the collections of cull Great Northern beans two to six random samples of 100 seeds showing evidence of bacterial infection were classified as to pathogen on the basis of the seed colorations listed previously. For the Pinto collections only the presence or absence of the pathogens among isolates from representative samples of infected seed was recorded. These data are summarized in Table 1.

All four pathogens were found generally and fairly uniformly distributed in collections of Great Northern cull beans from Nebraska and Wyoming in both years. Isolates from Wyoming and Colorado collections of cull Pinto beans included all but X. phaseoli var. fuscans. The failure to find this organism in Pinto beans may have resulted from its low incidence and the difficulty in recognizing its presence in pigmented seeds. This pathogen was found in garden beans grown in the Pinto-bean-producing area near Greeley, Colorado, in 1954, however.

The most prevalent pathogen in Great Northern samples was C. flaccumfaciens, which was responsible for the coloration in an average of 68 percent of the infected Great Northern seeds sampled in both years. The second in prevalence was C. flaccumfaciens var. auranti-acum, which infected 26 percent. This approached closely the 30 percent incidence reported for this pathogen in Nebraska cull collections in 1953 (3). X. phaseoli and X. phaseoli var. fuscans were found much less frequently, occurring in only 5 and 2 percent of the seeds, respectively.

In general, the orange wilt bacterium was more prevalent in collections from Scottsbluff, Gerring, and Minatare, in the eastern part of the Nebraska district than in those from Mitchell and Morril, Nebraska, and Torrington, Wyoming, to the west.

The 1954 collection from one of the Gerring establishments, which had a high percentage of orange-colored seeds, was from fields heavily damaged by hail. However, the suggestion that the orange variety is favored more than the yellow one by extensive hail damage seemed to be contradicted by the fact that the collection from Gerring in 1955 showing decidedly more orange than yellow seeds was from late-planted fields, which usually suffer much less hail damage than early-planted fields.

No consistent difference in distribution of the various organisms in electric-eye-sorted and gravity-and-hand-sorted collections was observable.

CONCLUSIONS AND DISCUSSION

Prevalence of the pathogens in the infected dry-bean seeds may or may not reflect the actual relative incidences of the respective diseases in the fields. It is probable that these causal bacteria differ in the extent to which they produce coloration in infected seeds. However, it is evident that the pathogens were present in all three districts. Their rather uniform general distribution in both years and among locations in Nebraska and Wyoming suggests that the pathogens are spread in wind-blown soil. Their relative incidences may be governed by their relative ability to survive in soil and be spread in this manner. In these wind-swept districts with many acres of light, sandy soils, this seems a logical explanation.

The wilt pathogens in dried infected seeds appeared to be more readily spread in water than the blight pathogens.

It seems likely that the characteristic of X. phaseoli var. fuscans responsible for darkened areas in infected seeds is the same as that causing darkening of media used for in vitro culturing of this bacterium (1).

Literature Cited

1. BURKHOLDER, W. H. 1930. The bacterial diseases of the bean, a comparative study. Cornell Univ. Agr. Exp. Sta. Mem. 127.
2. MAERZ, A. and M. REA PAUL. 1930. A dictionary of color. McGraw-Hill Book Co. Inc., New York.
3. SCHUSTER, M. L. and D. W. CHRISTIANSEN. 1953. An orange-colored bacterium comparable to Corynebacterium flaccumfaciens (Hedges) Dowson causing bean wilt. Phytopathology 43: 483.
4. SCHUSTER, M. L., and D. W. CHRISTIANSEN. 1957. An orange-colored strain of Corynebacterium flaccumfaciens causing bean wilt. Phytopathology 47: 51-53.

HORTICULTURAL CROPS RESEARCH BRANCH, AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE, IRRIGATION EXPERIMENT STATION, PROSSER, WASHINGTON

EFFECT OF BRUISING INJURY AND STORAGE TEMPERATURE UPON DECAY AND DISCOLORATION OF FRESH, IDAHO-GROWN ITALIAN PRUNES ON THE NEW YORK CITY MARKET

M. J. Ceponis and B. A. Friedman[1]

Recently, it has been claimed by some wholesale receivers at the New York City market that rough transit handling of fresh Idaho-grown Italian prunes was causing much bruising injury with a subsequent increase of decay and discoloration during the storage and marketing period. At their request samples from 61 one-half bushel baskets were removed during October 1956 from 39 rail shipments reported to have been handled roughly.

Each sample consisted of approximately 100 fruit chosen at random from each basket. The amount and type of decay was recorded, and the decayed fruit discarded. The remainder of the fruit was classified for bruising into four groups consisting of those showing no, slight, moderate, or severe bruising. Upon arrival at the laboratory approximately one-third of the prunes were cut and the amount of pulp (flesh) discoloration recorded as absent, slight, moderate, or severe. Another one-third was placed into bags and stored at 70° F for 3 to 5 days, and the remaining one-third stored at 35° for 14 days. After each storage period the decays were classified, and the sound fruit cut for determination of pulp discoloration.

Table 1. Relation of pulp discoloration to the amount of bruising injury of fresh, Idaho-grown Italian prunes.

Condition of fruit	Time of inspection	Number of fruit examined	Percent pulp discoloration			
			Slight	Moderate	Severe	Total
Unbruised	At arrival	914	11.2	0.8	0.1	12.1
	After 3-5 days at 70° F	845	45.2	17.5	14.4	77.1
	After 14 days 2t 35° F	905	37.2	1.8	0	39.0
Slightly bruised	At arrival	810	17.7	2.2	0.5	20.4
	After 3-5 days at 70° F	737	37.6	21.8	15.2	74.6
	After 14 days at 35° F	812	36.8	3.6	0.6	41.0
Moderately bruised	At arrival	202	25.3	7.9	2.0	35.2
	After 3-5 days at 70° F	176	40.3	21.6	13.6	75.5
	After 14 days at 35°F	190	46.3	11.1	1.6	59.0
Severely bruised	At arrival	60	40.0	15.0	13.3	68.3
	After 3-5 days at 70° F	35	40.0	31.4	20.0	91.4
	After 14 days at 35° F	47	36.2	23.4	6.4	66.0

At the arrival inspection, 6111 fruit were examined. Unbruised fruit amounted to 44.5 percent, slightly bruised 40.8 percent, moderately bruised 10.5 percent, and severely bruised 3.0 percent. Decayed fruit amounted to 1.2 percent. It should be noted that slight and moderate bruising would not ordinarily be considered to be of commercial significance, and by Federal standards only the severely bruised prunes would be graded as damaged.

The amount of pulp discoloration found at each inspection in each category of bruised fruit is shown in Table 1. As may be seen, the greater the severity of bruising, the greater the pulp discoloration. Pulp discoloration increased in storage, and was worse at 70° than at 35° F storage.

The relationship of bruising injury and storage temperature to the amount of decay is shown in Table 2. The results show that the greater the amount of bruising, the greater the percentage of decay. In addition, in all cases more decay developed at 70° than at 35° F storage.

A total of 379 decayed fruit was found in the three inspections. The type and percentage of decay are shown in Table 3. The blue mold fungus, Penicillium, which mainly enters fruit

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Table 2. Relationship of bruising injury and storage temperature to decay of fresh, Idaho-grown Italian prunes.

Condition of fruit	Duration and temperature of storage	Number of fruit examined	Amount of decay	
			Number fruit decayed	Percent decay
Unbruised	3-5 days at 70° F	915	76	8.3
	14 days at 35° F	918	12	1.3
Total		1833	88	4.8
Slightly bruised	3-5 days at 70° F	825	89	10.8
	14 days at 35° F	840	29	3.5
Total		1665	118	7.1
Moderately bruised	3-5 days at 70° F	216	39	18.1
	14 days at 35° F	211	19	9.0
Total		427	58	13.6
Severely bruised	3-5 days at 70° F	61	26	42.6
	14 days at 35° F	64	16	25.0
Total		125	42	33.6

Table 3. Types of decay found in fresh, Idaho-grown prunes upon arrival, and after storage for 3-5 days at 70° F or for 14 days at 35°, at New York City.

Type of decay	Incidence (percent)
Penicillium rot	45.6
Botrytis rot	24.3
Cladosporium rot	13.5
Penicillium rot plus Cladosporium rot	3.4
Penicillium rot plus Botrytis rot	2.8
Botrytis rot plus Cladosporium rot	2.6
Alternaria rot	2.1
Rhizopus rot	1.3
Cladosporium rot plus Alternaria rot	1.1
Stemphylium rot	1.1
Unidentified	1.1
Penicillium rot plus Rhizopus rot	0.5
Penicillium rot plus Alternaria rot	0.3
Penicillium rot plus Aspergillus rot	0.3

through injuries, accounted alone or in combination for 52.6 percent of the decay observed. There appears to be no previous report of the occurrence of Stemphylium (probably S. botryosum) or Aspergillus sp. on fresh prunes in this country.

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TWO PEPPERMINT DISEASES FOUND IN THE YAKIMA VALLEY OF WASHINGTON

C. B. Skotland and J. D. Menzies[1]

Verticillium Wilt: -- In 1955 a wilt disease of peppermint, Mentha piperita L., associated with Verticillium albo-atrum Reinke & Berth. was found causing considerable damage in a small area of one field in the Yakima Valley, Washington. At that time it was not known whether the pathogen was the mint strain (2) or a less pathogenic strain such as those previously reported in Oregon by Horner (1). In 1956 a further search for Verticillium wilt was made in this area. Isolations were made from many plants showing symptoms resembling wilt but Verticillium was rarely found except in the field previously mentioned. Four isolates were obtained from these collections; one was from the original infected field.

To test the pathogenicity of these isolates, mint cuttings with three pairs of leaves were dipped in a suspension of spores and microsclerotia and planted in a steamed sand-soil mixture. In 3 weeks, plants inoculated with two of the isolates were stunted and showed heavy pigmentation. After 5 weeks these plants were nearly dead and Verticillium could be isolated from all parts of the stem.

One of these isolates was from the originally discovered field and the other from a neighboring farm. The symptoms produced by the other two isolates at 3 weeks were a slight chlorosis and asymmetrical leaf development. In 5 weeks these plants could not be differentiated from the checks and Verticillium was isolated but rarely and from the lower stem only. However, those leaves which were deformed remained so. It is concluded that the two highly pathogenic isolates were probably the typical mint strain of Verticillium albo-atrum as found elsewhere. The others are relatively harmless strains. This is the first reported occurrence of this typical strain in Washington but Nelson (2) reported observing symptoms of Verticillium wilt in Washington in 1946.

Sclerotinia: -- In one of the fields in which Verticillium wilt was observed, Sclerotinia sclerotiorum (Lib.) DBy. emend Purdy was found attacking peppermint and causing considerable damage. The affected area was very small and was formerly a home garden. Extremely heavy mint growth favored the development of Sclerotinia. Abundant formation of sclerotia was found in the stem. No apothecia were observed. According to the "Index of Plant Disease in the United States" (3), this is the first report of the occurrence of this fungus on the genus Mentha. S. sclerotiorum has been found on one other species in the family Labiatae.

Literature Cited

1. HORNER, C. E. 1954. Pathogenicity of Verticillium isolates to peppermint. Phytopathology 44: 239-242.
2. NELSON, R. 1950. Verticillium wilt of peppermint. Michigan Agr. Exp. Sta. Tech. Bul. 221.
3. WEISS, F. and MURIEL J. O'BRIEN. 1952. Index of Plant Diseases in the United States. Plant Disease Survey Spec. Publ. No. 1, part IV.

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COLD INJURY TO YOUNG PINE TREES

George F. Weber

The fluctuation of temperatures in the Gainesville, Florida area from November 1956 through January 1957 is of considerable interest in regard to the type of damage that has been done to young trees. During late summer and early fall, before the latter part of November,

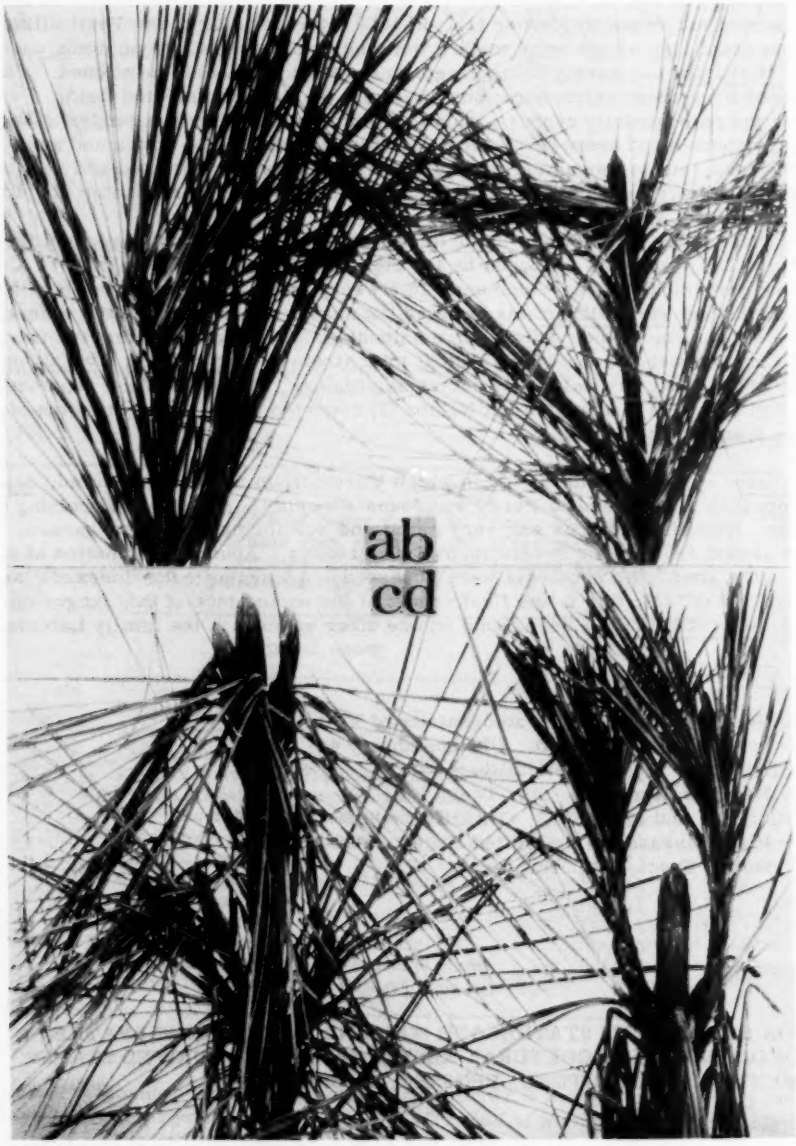

FIGURE 1. Symptoms of cold injury on terminal growth of pine trees were (a) the yellow area of the needles close to the fasicle; (b) more severe effects showed needles bent just above the fasicle sheath; (c) new growth and last flush needles killed; (d) brown and dead terminal shoots and killed needles.

the temperature in this region had not varied much from normal. On November 28, however, the official recorded temperature in Gainesville was 28° F, whereas a recording thermograph 2 1/2 miles from the official station indicated a temperature of 18° for over 2 hours' duration. About 1/2 mile from the official station, freezing temperatures did not occur, at least sufficient to freeze the foliage of poinsettia, hibiscus, or banana, all of which are considered tender plants. During December and the early part of January, for a period of about 7 weeks, temperatures ranged from occasional freezing minimums to daily maximums of 60° to 80°. Pine trees had begun to show new growth during this warm period, and by the middle of January were showing needle elongation at the fasicle sheath and flowers and terminal growth had started. These temperatures abruptly changed on January 19 when again freezing temperatures invaded the area, recording 20° F for a period of about 3 hours.

On February 20, 30 days after this second cold period, observations were made on several plantings of slash pine, Pinus elliotii. Small trees less than 6 feet high had shown little or no effect of the first freeze in November, and during December and the first part of January had made considerable new growth. On February 20 wide variation was evident in the damage resulting from the January cold period. Certain trees showed no effect of the cold at the growing tips, and in this case they were of a normal green color, and no damage apparently resulted. Trees that were slightly affected showed a definite yellowing of the needles at the growing tip, and these needles appeared to be most seriously affected at the extremity of the fasicle sheath (Fig. 1). From the end of the sheath the needles in some cases were yellow, and this yellow faded out to normal green toward the tip of the needle. Where the seedlings showed more severe effects, the area at the end of the sheaths was brown and apparently dead, followed by a definite yellowing of the needle which gradually faded out toward the tips. Where the damage was greater, browning of the needles was particularly severe; they were bent at the end of the sheath and hung down. On plants that had shown some new growth during this period, often an inch or two, the new growth was killed and was entirely brown and lifeless.

The severity of the effect apparently depended upon whether the plant had broken dormancy or not. All injury was confined to the growth produced during the last growing season, although in certain severe cases the needles of the previous year showed this same browning and drooping.

In pine plantings no apparent correlation between environmental factors and injury to the growing tips was noted. During the period indicated, from November 28 to February 20, there was little or no rain, and in many instances vegetation was showing effects of drouth conditions. Several inquiries have been made in the last week or two concerning this particular condition on slash pine seedlings by foresters in this part of the State.

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ANNOUNCEMENT

A "BIBLIOGRAPHY OF FOREST DISEASE RESEARCH IN THE DEPARTMENT OF AGRICULTURE", compiled by Agnes Ellis Moore, has just been published as United States Department of Agriculture Miscellaneous Publication No. 725. It can be obtained from the Superintendent of Documents, U. S. Government Printing Office, Washington 25, D. C. Price 55 cents.

More than 2200 references are listed alphabetically by authors. There is a very complete subject index.

A CORRECTION

REPORTER for March 15 (Vol. 41, No. 3), page 212: In the first sentence of the title for Figure 1, Phytophthora is misspelled.

(IN THIS ISSUE Continued).

G. R. DiMARCO and B. H. DAVIS tested several chemicals for effectiveness as post-harvest treatments to prevent decay of strawberries, page 460.

SAUL RICH and GORDON S. TAYLOR report results obtained with cottonseed oil formulations of organic fungicides applied to shade tobacco foliage, page 465.

V. R. WALLEN, J. K. RICHARDSON, L. CINQ-MARS, and W. BELL summarize results of the 1956 vegetable seed treatment trials, page 468.

EDWARD E. BUTLER reports that copper-treated fruit wraps did not reduce spread of Rhizopus rot in tomatoes, page 474.

JOHN L. LOCKWOOD, D. J. deZEEUW, A. L. ANDERSEN, and D. J. HAGEDORN summarize results of pea disease surveys in Michigan, page 478.

T. C. VANTERPOOL describes an abnormal type of inflorescence found on tomato, page 481.

D. W. BURKE and C. E. SELISKAR report that cultivator damage increased incidence of some stem and root diseases of bean, page 483.

DOUGLAS W. BURKE reports incidence of four bacterial pathogens in dry beans grown in irrigated districts of Nebraska, Wyoming, and Colorado, page 488.

M. J. CEPONIS and B. A. FRIEDMAN investigated factors resulting in decay and discoloration of fresh Idaho-grown prunes shipped to the New York City market, page 491.

C. B. SKOTLAND and J. D. MENZIES report occurrence of Verticillium wilt and Sclerotinia disease on peppermint in Washington, page 493.

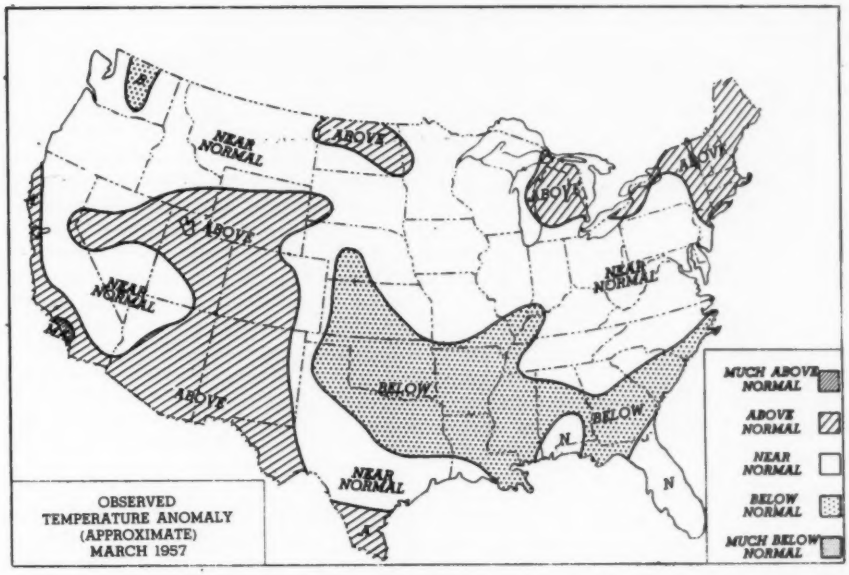
GEORGE F. WEBER describes symptoms on young slash pine trees in Florida, ascribed to cold injury, page 494.

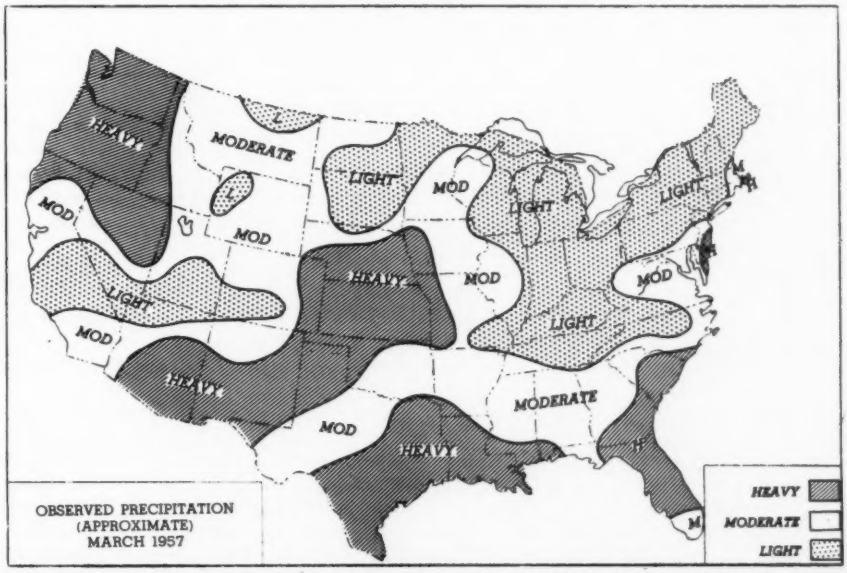
Announcement: Bibliography of Forest Disease Research, page 496.

Correction, page 496.

March weather, page 497.

Preparation of manuscripts for the Reporter, page 498.





The terms used in the accompanying maps, which define the ranges of temperature and precipitation, are numerical class limits. These are based on a statistical analysis of past records through which is determined the normal frequency of occurrence of temperatures and precipitation at various times of the year for different locations. For temperature the classes above, below, and near normal are so defined that they each normally occur one-fourth of the time; much above and much below normal, one-eighth of the time. Precipitation is depicted in terms of light, moderate, and heavy, each class normally occurring one-third of the time and thereby having equal probability of occurrence. These maps graphically represent only the general trends and give the country's weather picture at a glance. For quantitative studies, where monthly mean temperatures and actual precipitation records are needed for a given time and place, other publications of the Weather Bureau should be consulted. P. R. M.

MANUSCRIPTS FOR THE PLANT DISEASE REPORTER

(See also PDR 35: 121-122; 36: 214, 494)

(1) GENERAL: The Reporter page measures 9 inches long with the heading or 8 3/4 inches for the text part, by 6 inches wide. The copy is typed on a larger page, 11 1/4 inches of text or 12 inches overall in length by 8 inches in width, and reduced 25 percent in the photographic process of reproduction. Illustrations or tables larger in either dimension will take a correspondingly greater reduction. Only one size of type is available for text, footnotes, or tables.

(2) MANUSCRIPTS should be the original ribbon copy, not carbons, clearly typed and double-spaced throughout, including tables, footnotes, and bibliographies. (Note -- only one copy is needed.) Footnotes should be typed at the bottom of the page.

(3) ABSTRACTS are requested for all except very short articles. (PDR 38: 316).

(4) CAUSES OF DISEASES should be named. For bacteria, fungi, nematodes, etc., give the Latin name of the organism; for viruses either or both the accepted common name of the virus or a Latin name if you prefer it and there is one; for non-parasitic diseases state the causal factor if it is known. If the cause of a disease has not been determined say so. (PDR 36: 494).

(5) LITERATURE REFERENCES should be given in alphabetical order and numbered for citation in the text. We follow the AIBS suggestion of placing the year of publication after the author's name. Please check your titles carefully since we cannot do it for you. Be sure that text citations and bibliography agree; that foreign-language references are correct; that number or month is cited for periodicals that are not paged consecutively throughout the volume.

(6) NAMES OF FUNGICIDES should be given according to the suggestions that will be published in Phytopathology before long, and later repeated in the Reporter. In the meantime see PDR 35: 122.

(7) ILLUSTRATIONS should be sent to us unmounted. To prevent mistakes, write figure numbers on the back, and mark the top of each print when necessary. A sketch can show a preferred arrangement but please keep in mind page size, shape, and standard reduction (see above under General), and remember that figure titles and legends are part of the page. Lettering should be clear and large enough to be legible after reducing. Drawings, maps, and graphs can be photographs or originals, but should be finished and ready for reproduction, not just sketches.

(8) TABLES should be carefully thought out with particular attention to the Reporter's limitations in reproduction. Make titles and headings definite and self-explanatory. Designate footnotes in tables with superscript lower-case letters. Be sure that text discussion agrees with the data in the table. Do not abbreviate names of crop varieties.

(9) REPRINTS cannot be supplied since there is no way in which we can be reimbursed. However,

(10) The MULTILITH PLATES from which reprints can be made will be sent if requested at the time the article is submitted. The press size of these plates used for the Reporter is designated as small -- maximum image 9 1/2 by 13 inches, maximum paper size 9 3/4 by 14 inches -- for Model 1250. Most of the Experiment Stations have this type of multilith machine.

(11) We are the official correspondents in the United States of the PLANT PROTECTION BULLETIN published monthly by the United Nations Food and Agriculture Organization. We would appreciate it if plant pathologists would send us suitable material from time to time for inclusion in our monthly summary reports. Please mark it clearly so that we will know it is intended for the Bulletin. (See PDR 36: 492; 37: 637) -- P. R. M.

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